Attorney's Docket No. 046190/270365

PATENT

In The United States Patent and Trademark Office

In re: Sarem et al.
Appl No.: 10/692,529 Confirmation No.: 9904
Filed: Oct. 25, 2003 Group Art Unit:
For: A CELL AND TISSUE CULTURE DEVICE WITH TEMPERATURE REGULATION

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

STATEMENT OF FACTS IN SUPPORT OF FILING ON BEHALF OF NONSIGNING INVENTOR (37 C.F.R. § 1.47)

I, Daniel Ramey submit this statement in support of a petition under 37 C.F.R. § 1.47 to provide the facts that are relied upon to establish the diligent efforts made to secure the execution of the declaration by the nonsigning inventors for the above-identified patent application.

I am a patent attorney with the firm of Ernest Gutmann – Yves Plasseraud S.A., 3, rue Chauveau-Lagarde, Paris 75008 FRANCE. I represent the owner of this patent application, Societe Nouvelle Cell Tissue Progress, and I have knowledge of the facts recited herein.

The last known address of the non-signing inventors is as follows:

Farzin Sarem
34 Impasse des Azalees – L'Ile Verte, FR – 06560, Valbonne, France

Leila-Ouassila Sarem Damerdji
34 Impasse des Azalees – L'Ile Verte, FR – 06560, Valbonne, France

The facts of Mr. and Mrs. Sarem's refusal to sign a declaration for the present invention are provided as follows:

A. Background

Mr. Farzin Sarem and Mrs. Leila-Ouassila Sarem Damerdji (Mrs. Sarem) were employed by the French company Cell Tissue Progress beginning on February 25, 2000. A copy of the employment contracts of Mr. and Mrs. Sarem is enclosed. (Tab 1)

Mr. and Mrs. Sarem were listed as the inventors on French patent application No. 01.05651, which was filed on April 26, 2001, and assigned to Cell Tissue Progress. The present application claims priority from this French patent application.

The liquidation of Cell Tissue Progress was ordered in 2002 by the Court of Commerce of Paris, which appointed Mr. Denis Bouychou as curator. The Court of Commerce authorized the assignment of the assets of Cell Tissue Progress to Mr. Christopher Conway on March 12, 2002. A copy of this decision is attached. (Tab 2)

International application no. PCT/FR 02/01472 claiming priority from FR 01.10651 was filed on April 26, 2002, in the name of a new company founded by Mr. Conway called Societe Nouvelle Cell Tissue Progress. The assignment of FR 01.10651 and of its priority right to this new company was confirmed by an assignment deed executed by Mr. Bouychou on May 3, 2004. (Tab 3)

B. Inventors' Refusal to Sign Papers

When the international application no. PCT/FR 02/01472 was filed on April 26, 2002, my office sent a letter the same day to Mr. and Mrs. Sarem requesting that they sign a power of attorney form for the PCT application in addition to the corresponding declaration and assignment forms for the United States filing. When no response was forthcoming, the same letter was resent on June 20, 2002 by registered mail. The acknowledgment of receipt indicated that the inventors received this letter and the accompanying documents on June 24, 2002. Copies of these letters, along with the acknowledgment receipt of June 24, 2002, are attached. (Tab 4)

Mr. and Mrs. Sarem executed the PCT power of attorney form on July 24, 2002, and appointed the firm of Ernest Gutmann – Yves Plasseraud S.A to file the PCT Application for them and on their behalf. This PCT form was filed with the French Patent Office acting as the receiving office on August 1, 2002. The executed PCT power of attorney form and my related letter of August 1, 2002, to the French Patent Office are attached. (Tab 5)

I contacted Mr. Sarem personally by phone several times in 2002, and I have also spoken with a lawyer representing Mr. and Mrs. Sarem. Based on these conversations, it is my understanding that Mr. and Mrs. Sarem accept the fact that French patent application No. 01.10651 and all related rights belonged to Cell Tissue Progress, their former employer. However, even in light of the authorization of the assignment of rights in this invention by

the Court of Commerce, the inventors dispute the rights of Societe Nouvelle Cell Tissue Progress to their invention.

Accordingly, the Sarems refused to execute the corresponding U.S. declaration and assignment forms. As a result, my office sent new U.S. declaration and assignment forms and a copy of the corresponding U.S. application (Serial Number 10/692,529) to the inventors on October 21, 2004, by registered mail. The inventors received these documents on October 22, 2004. No reply has yet been received from the inventors. The letter and acknowledgment receipt are attached. (Tab 6)

C. Need to prevent irreparable damage or preserve the rights of the parties

A filing date is necessary to preserve the rights of the party having proprietary interest in this patent application and to prevent irreparable damage. A Notice to File Missing Parts under 35 U.S.C. § 371 was issued requiring filing of an oath or declaration of the inventors which must be submitted within two (2) months from the date of the date of the Notice to File Missing Parts. While this time period is temporarily extendable, Societe Nouvelle Cell Tissue Progress will suffer irreparable damage caused by continued fees to extend filing and eventual loss of priority date due to failure to file the missing parts within the prescribed statutory time period.

D. Statement of proprietary interest justifying taking action on behalf of inventors

At the time of the invention, the inventors were employed by Cell Tissue Progress and were obligated to assign their rights in the invention to Cell Tissue Progress. In addition, Mr. and Mrs. Sarem were listed as the inventors on the related French patent application No. 01.05651, which was filed on April 26, 2001, and assigned to Cell Tissue Progress.

The liquidation of Cell Tissue Progress was ordered in 2002 by the Court of Commerce of Paris, which appointed Mr. Denis Bouychou as curator. International application no. PCT/FR 02/01472 claiming priority from FR 01.10651 was filed on April 26, 2002, in the name of a new company founded by Mr. Christopher Conway called Societe Nouvelle Cell Tissue Progress. The assignment of FR 01.10651 and of its priority right to this new company was confirmed by an assignment deed executed by Mr. Bouychou on May 3, 2004. (Tab 3)

Thus, Societe Nouvelle Cell Tissue Progress has sufficient proprietary interest in the subject matter to justify the filing of this application on behalf of the inventors. The

accompanying declaration has been signed on behalf of the inventors by a duly authorized agent of Societe Nouvelle Cell Tissue Progress.

Daniel Ramey

Date: November 25, 2004

Daniel Ramey
Ernest Gutmann – Yves Plasseraud S.A.
3, rue Chauveau-Lagarde
Paris 75008
FRANCE

CONTRAT DE TRAVAIL

ENTRE LES SOUSSIGNES :

Cell Tissue Progress,
société anonyme au capital de 250 000 francs,
en cours d'immatriculation au RCS de Paris,
dont le siège social est situé au 8, bd Berthier, 75017 PARIS,

représentée par ***Monsieur Philibert du ROURE***, Président du Conseil d'Administration,
ayant tous pouvoirs à cet effet,

D'une part

Et

Monsieur Farzin SAREM
demeurant au 19, rue du Morvan, 54500 VANDOEUVRE LES NANCY,
né le 16 janvier 1953 à Téhéran (IRAN)
de nationalité française,

D'autre part

F. C.

IL A ETE CONVENU CE QUI SUIT :

ARTICLE 1 - ENGAGEMENT

Monsieur Farzin SAREM se déclare libre de tout engagement, et est engagé au sein de la société Cell Tissue Progress, sous réserve de la visite médicale d'embauche.

La Convention Collective applicable sera déterminée dans les deux mois de la création de la Société.

ARTICLE 2 - DUREE DU CONTRAT

Le présent contrat est conclu pour une durée indéterminée à compter du 25 février 2000, à neuf heures.

ARTICLE 3 - FONCTIONS

Monsieur Farzin SAREM est engagé en qualité de Manager Recherche et Développement, soumis au statut des cadres, responsable de la mise au point des produits et de la supervision de la production.

Ses fonctions ont, par nature, un caractère évolutif tenant, d'une part aux impératifs d'adaptation de l'entreprise et à ses besoins, d'autre part aux capacités et à l'approfondissement de la compétence de Monsieur Farzin SAREM.

Monsieur Farzin SAREM est rattaché à la Direction Générale.

Monsieur Farzin SAREM exercera ses fonctions de manière exclusive pour la société Cell Tissue Progress ou l'une de ses filiales.

ARTICLE 4 - LIEU DE TRAVAIL

Monsieur Farzin SAREM exercera ses fonctions à VANDOEUVRE LES NANCY dans un premier temps, puis au Centre de Recherche et Développement prévu d'être établi à Sophia Antipolis dans le courant de l'été 2000.

Pour faciliter le changement de domicile de Monsieur Farzin SAREM, il est entendu que celui-ci pourra bénéficier du remboursement de deux voyages (avec deux nuits d'hôtel chacun) dans la région de Sophia Antipolis et que ses frais de déménageurs (après approbation préalable du devis) lui seront également remboursés.

En fonction des nécessités de service, la société Cell Tissue Progress se réserve le droit de demander à Monsieur Farzin SAREM d'effectuer des déplacements temporaires. Monsieur Farzin SAREM sera alors remboursé de ses frais professionnels, conformément à l'article 7.

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ARTICLE 5 - DUREE DU TRAVAIL

Monsieur Farzin SAREM effectuera 39 heures par semaine, répartis sur les jours ouvrables. Cependant, compte tenu de la spécificité de ses fonctions, Monsieur SAREM en accord avec la Direction, bénéficiera de toute latitude pour adapter ses horaires aux nécessités de service.

ARTICLE 6 - REMUNERATION

En contrepartie de son travail, Monsieur Farzin SAREM percevra, pour sa première année de travail, une rémunération fixe annuelle brute de [redacted] francs.

Monsieur Farzin SAREM percevra une prime exceptionnelle de début d'activité de [redacted], qui sera payée avec le salaire de février 2000.

Il lui sera également attribuée une prime de fin d'année de 25% de son salaire fixe brut, soumise à la réalisation des ventes aux conditions du budget tel qu'approuvé par le Conseil d'Administration. Pour l'année 2000, la prime sera de [redacted] francs.

La rémunération globale de Monsieur Farzin SAREM (fixe & variable) sera redéfinie périodiquement par les organes dirigeants de la société Cell Tissue Progress. A cet effet, de façon formelle, Monsieur Farzin SAREM rencontrera son supérieur hiérarchique une fois l'an au minimum.

ARTICLE 7 - FRAIS PROFESSIONNELS

Les frais professionnels de Monsieur Farzin SAREM lui seront remboursés chaque mois sur présentation de justificatifs, dans le cadre d'une enveloppe globale définie par la Direction.

ARTICLE 8 : OBLIGATIONS PROFESSIONNELLES

Il est expressément convenu que Monsieur Farzin SAREM :

a) s'engage, pendant l'exécution du présent contrat, à consacrer toute son activité professionnelle et tous ses soins à la société Cell Tissue Progress ou à ses filiales. Il s'interdit, en conséquence, d'exercer une activité professionnelle soit pour son compte, soit pour le compte de tiers.

b) s'interdit expressément, pendant l'exécution du présent contrat, de s'intéresser directement ou indirectement, de quelque manière et à quelque titre que ce soit à toute affaire créée ou en voie de création, susceptible de faire concurrence à la société Cell Tissue Progress.

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c) s'engage à observer, pendant l'exécution du contrat, comme après sa rupture, la plus entière discrétion sur les méthodes et les activités de la société Cell Tissue Progress et de ses clients, ainsi qu'à ne lui délivrer aucun renseignement de nature à lui causer un préjudice.

L'inobservation de cette clause est susceptible d'être considérée comme étant constitutive d'une faute grave, voire lourde.

ARTICLE 9 - CONGES PAYES

Monsieur Farzin SAREM bénéficiera des avantages sociaux institués en vertu des dispositions légales, réglementaires et conventionnelles en faveur des cadres de la Société.

Monsieur Farzin SAREM bénéficiera des congés payés annuels attribués conformément aux lois, règlements et conventions en vigueur.

La période de ces congés est déterminée par accord entre la direction et Monsieur Farzin SAREM compte tenu des nécessités de service.

ARTICLE 10 - CONDITIONS D'EXECUTION DU CONTRAT

Monsieur Farzin SAREM s'engage à observer toutes les instructions et consignes particulières de travail qui lui seront données. Il devra également les transmettre à ses subordonnés et sera responsable de leur bonne application.

Monsieur Farzin SAREM devra faire connaître à l'entreprise sans délai toute modification postérieure à son engagement qui pourrait intervenir dans son état civil, sa situation de famille, son adresse.

ARTICLE 11 - CLAUSE DE NON-CONCURRENCE

Au cas où le présent contrat prendrait fin pour quelque cause que ce soit et quelle que soit la partie ayant pris l'initiative de la rupture, Monsieur Farzin SAREM s'interdit de s'intéresser, directement ou indirectement, à quelque titre ou de quelque manière que ce soit (salarié, non salarié, entreprise personnelle, associé, mandataire social, etc.) à toute entreprise créée ou en voie de création susceptible de concurrencer directement la société Cell Tissue Progress. Il est expressément convenu que l'exécution de la présente clause est limitée à une période de un an, à compter de la date de départ de Monsieur Farzin SAREM.

La société Cell Tissue Progress pourra libérer Monsieur Farzin SAREM de la clause de non-concurrence, à condition de le prévenir par écrit. Dans le cas où la clause de non concurrence serait appliquée, Monsieur Farzin SAREM aura droit à une indemnité selon les conditions prescrites par la Convention Collective applicable, ou, en cas d'absence dans celle-ci, selon les conditions prescrites par la Convention Collective la plus proche.

Après son départ de la société, Monsieur Farzin SAREM s'engage à respecter la plus stricte confidentialité sur toutes les informations qu'il détient concernant les activités de la société.

Au cas où Monsieur Farzin SAREM contreviendrait aux dispositions de la présente clause, il devrait verser à la société Cell Tissue Progress à titre d'indemnité forfaitaire, une somme égale au montant

F.C.

de la rémunération qu'il aurait acquise au titre de ses six derniers mois de rémunération par infraction constatée et ceci, indépendamment du droit qu'aurait la société Cell Tissue Progress de faire cesser cette contravention par toutes les voies du droit.

Le présent contrat est établi en deux exemplaires originaux.

Fait à Paris, le 22 février 2000

Pour Cell Tissue Progress,
Monsieur Philibert du ROURE

Monsieur Farzin SAREM

CONTRAT DE TRAVAIL

ENTRE LES SOUSSIGNES :

Cell Tissue Progress,
société anonyme au capital de 250 000 francs,
en cours d'immatriculation au RCS de Paris,
dont le siège social est situé au 8, bd Berthier, 75017 Paris

représentée par ***Monsieur Philibert du ROURE***, Président du Conseil d'Administration,
ayant tous pouvoirs à cet effet,

D'une part

Et

Madame Leila SAREM
demeurant 19, rue du Morvan, 54500 VANDOEUVRE LES NANCY
née le 20 mai 1958 à Alger (ALGERIE)
de nationalité française,

D'autre part

LS

IL A ETE CONVENU CE QUI SUIT :

ARTICLE 1 - ENGAGEMENT

Madame Leila SAREM se déclare libre de tout engagement, et est engagée au sein de Cell Tissue Progress, sous réserve de la visite médicale d'embauche.

La Convention Collective applicable sera déterminée dans les deux mois de la création de la Société.

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Madame Leila SAREM est engagée en qualité de Responsable du Contrôle de la Qualité des Produits et Biologie, soumise au statut des Cadres.

Ses fonctions ont, par nature, un caractère évolutif tenant, d'une part aux impératifs d'adaptation de l'entreprise et à ses besoins, d'autre part aux capacités et à l'approfondissement de la compétence de Madame Leila SAREM.

Madame Leila SAREM est rattachée à la Direction Générale.

Madame Leila SAREM exercera ses fonctions de manière exclusive pour la société Cell Tissue Progress ou l'une de ses filiales.

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Madame Leila SAREM exercera ses fonctions à VANDOEUVRE LES NANCY dans un premier temps, puis au Centre de Recherche et Développement prévu d'être établi à Sophia Antipolis, dans le courant de l'été 2000.

Pour faciliter le changement de domicile de Madame Leila SAREM, il est entendu que Madame Leila SAREM pourra bénéficier du remboursement de deux voyages (avec deux nuits d'hôtel chacun) dans la région de Sophia Antipolis et que ses frais de déménageurs (après approbation préalable du devis) lui seront également remboursés.

En fonction des nécessités de service, la société Cell Tissue Progress se réserve le droit de demander à Madame Leila SAREM d'effectuer des déplacements temporaires. Madame Leila SAREM sera alors remboursée de ses frais professionnels, conformément à l'article 7.

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Madame Leila SAREM effectuera 39 heures par semaine, répartis sur les jours ouvrables. Cependant, compte tenu de la spécificité de ses fonctions, Madame Leila SAREM, en accord avec la Direction, bénéficiera de toute latitude pour adapter ses horaires aux nécessités de service.

ARTICLE 6 - REMUNERATION

En contrepartie de son travail, Madame Leila SAREM percevra, pour sa première année de travail, une rémunération fixe annuelle brute de [redacted] francs.

Madame Leila SAREM percevra une prime exceptionnelle de début d'activité de [redacted] qui sera payée avec le salaire de février 2000.

Il lui sera également attribuée une prime de fin d'année de 10% du salaire brut, soumise à la réalisation des ventes aux conditions du budget tel qu'approuvé par le Conseil d'Administration. Pour l'année 2000, la prime sera de [redacted] francs.

La rémunération de Madame Leila SAREM (fixe + variable) sera redéfinie périodiquement par les organes dirigeants de la société Cell Tissue Progress. A cet effet, de façon formelle, Madame Leila SAREM rencontrera son supérieur hiérarchique une fois l'an au minimum.

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Les frais professionnels de Madame Leila SAREM lui seront remboursés chaque mois sur présentation de justificatifs, dans le cadre d'une enveloppe globale définie ultérieurement par la Direction.

ARTICLE 8 : OBLIGATIONS PROFESSIONNELLES

Il est expressément convenu que Madame Leila SAREM :

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b) s'interdit expressément, pendant l'exécution du présent contrat, de s'intéresser directement ou indirectement, de quelque manière et à quelque titre que ce soit à toute affaire créée ou en voie de création, susceptible de faire concurrence à la société Cell Tissue Progress.

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c) s'engage à observer, pendant l'exécution du contrat, comme après sa rupture, la plus entière discrétion sur les méthodes et les activités de la société Cell Tissue Progress et de ses clients, ainsi qu'à ne lui délivrer aucun renseignement de nature à lui causer un préjudice.

L'inobservation de cette clause est susceptible d'être considérée comme étant constitutive d'une faute grave, voire lourde.

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Madame Leila SAREM bénéficiera des congés payés annuels attribués conformément aux lois, règlements et conventions en vigueur.

La période de ces congés est déterminée par accord entre la direction et Madame Leila SAREM compte tenu des nécessités de service.

ARTICLE 10 - CONDITIONS D'EXECUTION DU CONTRAT

Madame Leila SAREM s'engage à observer toutes les instructions et consignes particulières de travail qui lui seront données. Elle devra également les transmettre à ses subordonnés et sera responsable de leur bonne application.

Madame Leila SAREM devra faire connaître à l'entreprise sans délai toute modification postérieure à son engagement qui pourrait intervenir dans son état civil, sa situation de famille, son adresse.

ARTICLE 11 - CLAUSE DE NON-CONCURRENCE

Au cas où le présent contrat prendrait fin pour quelque cause que ce soit et quelle que soit la partie ayant pris l'initiative de la rupture, Madame Leila SAREM s'interdit de s'intéresser, directement ou indirectement, à quelque titre ou de quelque manière que ce soit (salarié, non salarié, entreprise personnelle, associé, mandataire social, etc.) à toute entreprise créée ou en voie de création susceptible de concurrencer directement la société Cell Tissue Progress. Il est expressément convenu que l'exécution de la présente clause est limitée à une période de un an, à compter de la date de départ de Madame Leila SAREM.

La société Cell Tissue Progress pourra libérer Madame Leila SAREM de la clause de non-concurrence, à condition de la prévenir par écrit. Dans le cas où la clause serait appliquée, Madame Leila SAREM aura droit à une indemnité selon les conditions prescrites par la Convention Collective applicable, ou, en cas d'absence dans celle-ci, selon les conditions prescrites par la Convention Collective la plus proche.

Après son départ de la société, Madame Leila SAREM s'engage à respecter la plus stricte confidentialité sur toutes les informations qu'elle détient concernant les activités de la société.

L.S

Au cas où Madame Leila SAREM contreviendrait aux dispositions de la présente clause, elle devrait verser à la société Cell Tissue Progress à titre d'indemnité forfaitaire, une somme égale au montant de la rémunération qu'elle aurait acquise au titre de ses six derniers mois de rémunération par infraction constatée et ceci, indépendamment du droit qu'aurait la société Cell Tissue Progress de faire cesser cette contravention par toutes les voies du droit.

Le présent contrat est établi en deux exemplaires originaux.

Fait à Paris, le 22 février 2000

Pour Cell Tissue Progress,
Monsieur Philibert du ROURE

Madame Leila SAREM

ORDONNANCE

Nous,

Président du Tribunal de commerce de Paris,

Vu les articles L.237-6 du Code de commerce et 875 du Nouveau code de procédure civile,
Vu la requête qui précède et les pièces à l'appui, notamment la proposition de reprise par Monsieur Conway qui restera annexée à la présente requête.
Vu l'urgence,

- Constatons que tous les actionnaires, ainsi que le liquidateur et le commissaire aux comptes de la société CELL TISSUE PROGRESS, ont été dûment consultés sur la proposition de reprise partielle d'actifs présentée par Monsieur John Christopher Conway ;

- Constatons que la majorité des actionnaires en nombre et en voix de ladite société et le liquidateur ont approuvé le projet de reprise ; que le commissaire aux comptes ne s'y oppose pas.

- Constatons néanmoins que l'unanimité des actionnaires, exigée du requérant en raison de ses qualités d'actionnaire et d'ancien dirigeant de la société CELL TISSUE PROGRESS, n'est pas acquise.

<u>En conséquence,</u>

- Autorisons la cession partielle d'actifs de la société CELL TISSUE PROGRESS dans les termes de la proposition présentée par Monsieur John Christopher Conway aux actionnaires, au liquidateur et au commissaire aux comptes de ladite société.

- Disons qu'il nous en sera référé en cas de difficulté.

- Disons la présente décision exécutoire sur minute, nonobstant l'exercice de toute voie de recours.

Donnée à Paris, le 19 3 2002

Pour le Président
Le Président de Chambre Délégué

F. CAMBOURNAC

TRIBUNAL DE COMMERCE DE PARIS

LE GREFFIER

CESSION
ASSIGNMENT

Le soussigné: **Denis BOUYCHOU, Administrateur Judiciaire**
I/We, de la Société CELL TISSUE PROGRESS, Société Anonyme
dont le siège social est 8 boulevard Berthier, 75017 Paris, France

*Curator of the « **CELL TISSUE PROGRESS**", a stock company*
Having a registered office at 8 boulevard Berthier, 75017 Paris France

déclare céder à:
declare that I/we assign to:

SOCIETE NOUVELLE CELL TISSUE PROGRESS
société à responsabilité limitée
dont le siège social est 40 rue Damrémont, 75018 Paris, France,
représentée par Mr. **John Christopher CONWAY**
son Gérant,

SOCIETE NOUVELLE CELL TISSUE PROGRESS
a limited liability company
having a registered office at 40 rue Damrémont, 75018 Paris, France,
*represented by Mr. **John Christopher CONWAY***
its manager

Tous les droits, y compris le droit de priorité, relatifs aux brevets, demandes de brevet et marques déposées cités à l'annexe 1.

All the rights, including the priority right, on the patents, patent applications and trademarks listed on appendix 1.

Fait le 3 mars 2009 à Paris, France,

Dated this *in*

Denis BOUYCHOU
Administrateur Judiciaire

Denis BOUYCHOU
Administrateur Judiciaire
95, rue Saint-Lazare
75009 PARIS

Témoins : Laurent BARBE
Witnesses : Daniel Ramey

3 rue Chanveau - Lagarde
F- 75008 Paris

ANNEXE 1

Nos Réf.	Titre du brevet, du dossier ou de la marque	Date Dépôt	Numéro Dépôt	Date Acc. ou Enreg.	Numéro Acc. ou Enreg.	Titulaire	Pays	Nature	Etat	Réf. client
B4472	ROBOT DE CULTURE DE CELLULES ET DE TISSUS	17/01/2000	0000548			CELL TISSUE PROGRESS	FRANCE	BREVET D'INVENTION	EN VIGUEUR	
B4472A	ROBOT DE CULTURE DE CELLULES ET DE TISSUS	20/09/2000	09/666,592	07/08/2001	6271027	CELL TISSUE PROGRESS	ETATS UNIS	BREVET D'INVENTION	EN VIGUEUR	
B4472B	ROBOT DE CULTURE DE CELLULES ET DE TISSUS	15/01/2001	PCT/FR 01/00121			CELL TISSUE PROGRESS	P.C.T.	BREVET D'INVENTION	EN VIGUEUR	
B4472BA	ROBOT DE CULTURE DE CELLULES ET DE TISSUS	15/01/2001	2,407,012			CELL TISSUE PROGRESS	CANADA	BREVET D'INVENTION	EN VIGUEUR	
B4472BB	ROBOT DE CULTURE DE CELLULES ET DE TISSUS	15/01/2001	2001-553910			CELL TISSUE PROGRESS	JAPON	BREVET D'INVENTION	EN VIGUEUR	
B4472C	ROBOT DE CULTURE DE CELLULES ET DE TISSUS	15/01/2001	01400108.5			CELL TISSUE PROGRESS	EUROPE	BREVET D'INVENTION	EN VIGUEUR	
B4569	UNITE DE CULTURE DE CELLULES ET TISSUS A CONFIGURATION VARIABLE	27/04/2000	0005413			CELL TISSUE PROGRESS	FRANCE	BREVET D'INVENTION	EN VIGUEUR	
B4569A	UNITE DE CULTURE DE CELLULES ET TISSUS A CONFIGURATION VARIABLE	20/09/2000	09/665,704	05/11/2002	6,475,777	CELL TISSUE PROGRESS	ETATS UNIS	BREVET D'INVENTION	EN VIGUEUR	
B4569B	UNITE DE CULTURE DE CELLULES ET TISSUS A CONFIGURATION VARIABLE	27/04/2001	PCT/FR 01/01330			CELL TISSUE PROGRESS	P.C.T.	BREVET D'INVENTION	EN VIGUEUR	
B4800	DISPOSITIF DE CULTURE CELLULAIRE ET TISSULAIRE A REGULATION ...	26/04/2001	0105651	01/08/2003	0105651	CELL TISSUE PROGRESS	FRANCE	BREVET D'INVENTION	EN VIGUEUR	

Denis BOUYCHOU
Administrateur Judiciaire
95, rue Saint-Lazare
75009 PARIS

ERNEST GUTMANN - YVES PLASSERAUD S.A.
SOCIETE DE CONSEILS EN PROPRIETE INDUSTRIELLE

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20. 6. 2002

Monsieur et Madame SAREM
34 Impasse des Azalées
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FRANCE

VOTRE REFERENCE :

NOTRE REFERENCE : B4800 – CM/SZ

Paris, 26 avril 2002

"DISPOSITIF DE CULTURE CELLULAIRE ET TISSULAIRE A REGULATION ..." – Invention : SAREM DAMERDJI LEILA; SAREM FARZIN

Extension à l'étranger de la demande de brevet France n° 0105651 déposée le 26 avril 2001 au nom de CELL TISSUE PROGRESS

Madame, Monsieur,

L'administrateur judiciaire chargé de la liquidation de la société Cell Tissue Progress a cédé les droits sur la demande de brevet français citée en référence à la Société Nouvelle Cell Tissue Progress, laquelle nous a chargés de procéder au dépôt d'une demande internationale PCT revendiquant la priorité de la demande de brevet français précitée.

Pour cela, nous devons déposer un pouvoir de mandataire signé par vous-mêmes en tant qu'inventeurs désignés dans la demande de brevet français.

Nous joignons ce pouvoir en annexe et vous prions de nous le retourner signé aux endroits indiqués, avant le **15 mai 2002.**

Nous joignons également une déclaration et une cession, que nous devrons déposer aux USA et que nous vous prions de nous retourner après les avoir signées, toujours en votre qualité d'inventeurs.

Nous sommes bien entendu à votre disposition pour toute information complémentaire que vous souhaiteriez nous demander.

Nous vous prions d'agréer, Madame, Monsieur, nos salutations distinguées.

Daniel Ramey

PJ :

LA POSTE

AVIS DE RÉCEPTION DE VOTRE ENVOI RECOMMANDÉ

RA 1022 5881 5FR

AR/CM B 4800

1[er]

VALBONNE CC 24-6 2002 06 ALPES-MMES

Présenté le : 24/6/02

Distribué le :

Signature du destinataire

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DUPLICATA

I.N.P.I.
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F-75008 PARIS

A l'attention du service des brevets PCT

VOTRE REFERENCE : B4800A-DR/MCT/LB

NOTRE REFERENCE : Paris, 01 août 2002

Demande de brevet Internationale n° PCT/FR02/01472 déposée le 26 avril 2002 au nom de SOCIETE NOUVELLE DU PELL TISSUE PROGRESS

INPI
- 2 AOUT 2002

Messieurs,

Afin de régulariser le dossier de la demande de brevet ci-dessus mentionnée, nous vous prions de trouver les documents suivants qui n'étaient pas joints au moment du dépôt :

- Pouvoir

Nous vous remercions de nous retourner le duplicata de la présente muni de votre cachet

et vous prions d'agréer, Messieurs, l'expression de notre considération distinguée.

Marie-Christine TRUONG-VINH-TONG
Service Dépôts Brevets

PJ : ci-dessus mentionnées.

BREVETS
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(1)Agence de Lyon
(2)Agence d'Alicante

SOCIETE ANONYME
AU CAPITAL DE 500 000 €
RCS PARIS B 332 417 500
APE 741 A

POUVOIR
POWER

Le(s) soussigné(s) : 1) SAREM Farzin — 2) SAREM DAMERDJI Leila-Ouassila
the undersigned:

demeurant à : 34, Impasse des Azalées, Ile Verte, 06560 Valbonne, France
residing at: 34, Impasse des Azalées, Ile Verte, 06560 Valbonne, France

donne(nt) par le présent pouvoir, à :
hereby appoints and gives power to:

ernest gutmann - yves plasseraud s.a.
3 rue Chauveau Lagarde - F-75008 PARIS (FRANCE)

Pour eux et en leurs noms, déposer une demande internationale PCT (pour les Etats-Unis d'Amérique) pour :
For them and on their behalf, to file an international application PCT (for the USA) for:

"Dispositif de culture cellulaire et tissulaire à régulation thermique"

en conséquence, verser, le montant des taxes, signer toutes pièces et registres, donner quittance, présenter toutes demandes, y inclus des demandes relatives à des additions, lever l'expédition des titres ou certificats officiels, retirer les pièces et les taxes en cas de rejet ou de retrait des demandes, présenter toutes demandes d'autorisation, d'introduire des modèles, prendre la parole, élire domicile, substituer au besoin pour tout ou partie de l'exécution du présent mandat et, généralement remplir toutes les formalités voulues par les lois, déclarant reconnaître et ratifier tous les actes accomplis pour la réalisation du présent mandat.

accordingly, to pay all taxes, to sign all documents and registers, to deliver discharges, to file all applications, including applications for additions, improvements, extensions of terms, claims or rectifications, to receive all official certificates or titles, to withdraw all documents and recover all taxes in case of rejection or withdrawal, to introduce request for authorizations, to provide samples, to make declarations, to establish residence, to sub-delegate if required all or part of the present power and, in general, to accomplish all formal acts required by statutes, and hereby declares that all acts achieved with a view of fulfilling the present mandate will be recognized and ratified.

Fait à Valbonne le 24/07/2002 — Signed at Valbonne on 24/07/2002.

Leila SAREM

Farzin Sarem

- Indiquer la qualité du signataire
- Mentionner en entier le prénom et le nom
- Pas d'attestation de signature
- Pas de légalisation

- Write down full first name and surname
- Indicate the business capacity of the signatories
- Attestation of signature not required
- No legalization required

LA POSTE

AVIS DE RÉCEPTION DE VOTRE ENVOI RECOMMANDÉ

RA 2946 5744 8FR

B 4800 A1 - JR / Ade. 6

VALBONNE-CC.11 * 22 10 2004 * 90-ALPES MMES

Présenté le : 22/10

Distribué le :

Signature du destinataire :

M^r & M^me SAREM

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Recommandé A. R.

Monsieur et Madame SAREM
34 Impasse des Azalées – l'Ile Verte
FR-06560 VALBONNE

VOTRE REFERENCE :

NOTRE REFERENCE : B4800AA-DR/PBE

Paris, 21 octobre 2004

"THERMALLY OPERATED CELL AND TISSUE CULTURE DEVICE"
Invention : SAREM DAMERDJI LEILA; SAREM FARZIN
ETATS UNIS - Demande de brevet n° 10/692,529 déposée le 24 octobre 2003
au nom de SOCIETE NOUVELLE CELL TISSUE PROGRESS

Chère Madame, Cher Monsieur,

Dans le cadre de la demande de brevet mentionnée ci-dessus, nous vous prions de bien vouloir trouver ci-joint :

- 1 Déclaration,
- 1 Cession,

que nous vous demandons de bien vouloir nous retourner, dûment signées par vos soins, de préférence avant le 5 novembre 2004.

Nous vous en remercions vivement et,

Vous prions d'agréer, chère Madame, cher Monsieur, nos salutations les meilleures.

Monique EPINEAU
Service Dépôt Brevets

P.J. : 2

SOCIETE ANONYME
AU CAPITAL DE 500 000 €
RCS PARIS B 332 417 500
APE 741 A

ASSIGNMENT - WORLDWIDE

For good and valuable consideration, the receipt and sufficiency of which are hereby acknowledged, each undersigned inventor has sold and assigned, and by these presents hereby sells and assigns, unto

SOCIETE NOUVELLE CELL TISSUE PROGRESS
40 rue Danremont
FR-75018 Paris
FRANCE

its successors and assigns, the entire right, title and interest, so far as concerns the United States and the Territories and Possessions thereof in and to the invention in **"A CELL AND TISSUE CULTURE DEVICE WITH TEMPERATURE REGULATION"**

as set forth in this United States Patent Application

- ☐ executed concurrently herewith
- ☐ executed on _____
- ☒ Application No. 10/692,529; filed October 24, 2003
- ☒ Application claims priority from PCT Application No. PCT/FR02/01472, filed April 26, 2002, which claims French priority from Application No. 0105651, filed April 26, 2001

all applications listed above being hereinafter referred to as the "application(s)";

said application for United States Letters Patent, including all divisional, renewal, substitute, continuation, nonprovisionals, continuation-in-parts, and Convention applications based in whole or in part upon said inventions or upon said applications, and any and all Letters Patent and reissues, reexaminations, and extensions of Letters Patent granted for said inventions or upon said applications and every priority right that is or may be predicated upon or arise from said inventions, said applications, and said Letters Patent; said Assignee being hereby authorized to file patent applications in any or all countries on any or all said inventions in the name of the undersigned or in the name of said Assignee or otherwise as said Assignee may deem advisable, under the International Convention or otherwise; the Commissioner of Patents and Trademarks of the United States of America being hereby authorized to issue or transfer all said Letters Patent to said Assignee in accordance herewith; this assignment being under covenant, not only that full power to make the same is had by the undersigned, but also that such assigned right is not encumbered by any grant, license, or other right theretofore given, and that the undersigned will do all acts reasonably serving to ensure that the said inventions, patent applications and Letters Patent shall be held and enjoyed by said Assignee as fully and entirely as the same could have been held and enjoyed by the undersigned if this assignment had not been made, and particularly to execute and deliver to said Assignee all lawful documents including petitions, specifications, oaths, assignments, invention disclaimers, declarations, and lawful affidavits in form and substance which may be requested by said Assignee, to furnish said Assignee with all facts relating to said inventions or the history thereof and any and all documents, photographs,

models, samples or other physical exhibits which may embody said inventions, and to testify in any proceedings relating to said inventions, patent applications, and/or Letters Patent.

The undersigned hereby grant(s) an authorized representative of Assignee the power to insert in this Assignment any further identification that may be necessary or desirable to comply with the rules of the U.S. Patent and Trademark Office for recordation of this Assignment.

_____ _____

Date **Farzin Sarem**

_____

Witness

_____

Witness

_____ _____

Date **Leila-Ouassila Sarem Damerdji**

_____

Witness

_____

Witness

Attorney Docket No. 046190/270365

Attorney Docket No. 046190/270365

DECLARATION AND POWER OF ATTORNEY FOR PATENT APPLICATION

As a below named inventor, I hereby declare that:

My residence, mailing address and citizenship are as stated below next to my name.

I believe I am the original, first and sole inventor (if only one name is listed below) or an original, first and joint inventor (if plural names are listed below) of the subject matter which is claimed and for which a patent is sought on the invention entitled

A CELL AND TISSUE CULTURE DEVICE WITH TEMPERATURE REGULATION,

the specification of which

☐ is attached hereto

OR

☒ was filed on October 24, 2003 as United States Application No. 10/692,529, which was filed as a continuation application of PCT/FR02/01472, filed April 26, 2002.

I hereby state that I have reviewed and understand the contents of the above-identified specification, including the claims, as amended by any amendment referred to above.

I acknowledge the duty to disclose information which is material to patentability as defined in 37 CFR 1.56, including for continuation-in-part applications, material information which became available between the filing date of the prior application and the national or PCT international filing date of the continuation-in-part application.

I hereby claim foreign priority benefits under 35 U.S.C. 119(a)-(d) or (f), or 365(b) of any foreign application(s) for patent, inventor's or plant breeder's rights certificate(s), or 365(a) of any PCT international application which designated at least one country other than the United States of America, listed below and have also identified below, by checking the box, any foreign application for patent, inventor's or plant breeder's rights certificate(s), or any PCT International application having a filing date before that of the application on which priority is claimed.

Prior Foreign Application Number(s)	Country	Foreign Filing Date (MM/DD/YYYY)	Priority Not Claimed	Certified Copy Attached?	
				Yes	No
FR 01/05651	France	04/26/2001	☐	☐	☒

I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under 18 U.S.C. 1001 and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

The undersigned hereby authorizes the U.S. attorney or agent named herein to accept and follow instructions from my French representatives, Ernest Gutmann - Yves Plasseraud SA, as to any action to be taken in the U.S. Patent and Trademark Office regarding this application without direct communication between the U.S. attorney or agent and the undersigned. In the event of a change in the persons from whom instructions may be taken, the U.S. attorney or agent named herein will be so notified by the undersigned.

POWER OF ATTORNEY: As a named inventor, I hereby appoint the practitioners associated with the Customer Number provided below to prosecute this application and to transact all business in the Patent and Trademark Office connected therewith, and direct that all correspondence be addressed to that Customer Number:

Customer Number 00826

Direct telephone calls to: Raymond O. Linker, Jr.
Registration No. 26,419

Tel Charlotte Office (704) 444-1000
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Full name of first (sole) inventor: **Farzin Sarem**

Inventor's Signature: _____ Date: _____
Residence: Valbonne, France
Citizenship: France
Mailing Address: 34 Impasse des Azalees - I'lle Verte
FR-06560
Valbonne, France

Full name of second inventor: **Leila-Ouassila Sarem Damerdji**

Inventor's Signature: _____ Date: _____
Residence: Valbonne, France
Citizenship: France
Mailing Address: 34 Impasse des Azalees - I'lle Verte
FR-06560
Valbonne, France

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Application of: Farzin SAREM
Leila-Ouassila SAREM DAMERDJI

Serial No:

Filed:

For: A CELL AND TISSUE CULTURE DEVICE WITH TEMPERATURE REGULATION

DECLARATION

I, Andrew Scott Marland, of 35, avenue Chevreul, 92270 BOIS COLOMBES, France, declare that I am well acquainted with the English and French languages and that the attached translation of the French language PCT international application, Serial No. **PCT/FR02/01472** is a true and faithful translation of that document as filed.

All statements made herein are to my own knowledge true, and all statements made on information and belief are believed to be true; and further, these statements are made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any document or any registration resulting therefrom.

Date: October 14, 2003

Andrew Scott Marland

A CELL AND TISSUE CULTURE DEVICE WITH TEMPERATURE REGULATION

The present invention relates to the field of dynamically culturing cells and tissue using a culture fluid or nutrient medium set into motion.

The invention relates more precisely to devices for culturing cells and tissue comprising: i) one or more culture wells defining chambers for receiving cells or tissue to be cultured; ii) first and second reservoirs each housing at least one flexible bag, at least one of which is suitable for receiving a culture fluid; iii) link means coupled to the well(s) and to the bags in order to enable culture fluid to flow from one reservoir to the other via the well(s); and iv) pressurization means enabling the bags of the first and second reservoirs to be subjected respectively to first and/or second sequences of external pressures which are defined by one or more control modules and which serve to govern the flow of culture fluid in the well(s).

That type of device, as described in French patent application No. 00/00548, enables suitable flow conditions to be maintained throughout the duration of culturing. However, when the culture requires an environment that is under temperature control, devices of that type need to be placed inside a suitable incubator, thereby increasing the biological risks associated with displacements, costs, handling, and size, and makes it impossible to use a microscope to observe how the culture is progressing while incubation is taking place. In addition, the transfers lead to temperature changes that can give rise to harmful biological consequences.

An object of the invention is to provide an original solution to all or some of the above-specified drawbacks.

To this end, the invention provides a device of the type described in the introduction, in which temperature regulation means under the control of a control module

are provided and serve to maintain a first selected temperature or a first selected temperature cycle within the well(s) and/or to apply a second selected temperature or a second selected temperature cycle to the culture fluid leaving at least one of the first and second reservoirs in order to feed the well(s).

Thus, temperature regulation within the device can be performed either exclusively at well level, or else exclusively at the level of the culture fluid feeding the wells, or indeed simultaneously both at well level and at the level of the culture fluid so as to minimize temperature disturbances when making exchanges between the culture fluid and the cells.

The first and second temperatures (or the first and second temperature cycles) are selected as a function of the type of culture. They can therefore be substantially identical, or else they can be different if so required by the culture. The second temperatures (or the second cycles) can also vary from one reservoir to the other should that be necessary. It is also possible to vary the temperatures (or the cycles) during the progress of culturing. To do this, parameters for causing temperatures to vary may be programmed, e.g. by being included in the program that determines the external pressure sequences imparted by the control module. Such programming may be performed using an input interface, or else directly by transferring predefined programs into a memory of the device that is coupled with (or integrated in) the control module, and then selecting one of the programs (the memory may optionally be re-writable via the above-mentioned interface).

In a first embodiment of the device of the invention, the temperature regulation means comprise a heating fluid circuit, the circuit comprising at least a first portion integrated in the walls defining the well(s) (possibly in the form of flow channels formed at the periphery of the chambers, or spaces for allowing

fluid flow formed in the walls of the wells and connected to first connection means), or second and third portions integrated respectively in the walls defining the first and second reservoirs and arranged to enable a heat-conveying fluid to flow (these might be spaces formed between an inner shell and an outer shell which, once assembled together, define the first and second reservoirs), or else a combination of the first, second, and third portions. In the combination case, the second portion of the fluid circuit is preferably arranged to feed the heat-conveying fluid (liquid or gas) to the first portion, while the third portion is arranged to select the heat-conveying fluid that has flowed through the first portion. It is then particularly advantageous for the second and third portions of the fluid circuit to include second and third connection means opening out into the space between shells and suitable for being connected respectively to the first connection means and to a (main) fourth portion of the fluid circuit for feeding and collecting the heat-conveying fluid.

In this first embodiment, the heating fluid circuit preferably includes a pump coupled to a main container containing a fraction of the heat-conveying fluid (liquid or gas) and electric heater means (such as heater resistances, for example) for heating the heat-conveying fluid in controlled manner before it feeds the first, second, and third portions.

In a second embodiment of the device of the invention, the temperature regulation means comprise either first electric heater elements for providing at least a portion of the controlled heating of the well (e.g. in the form of heater resistances placed against or insulated in the walls of the wells), or else second electric heater elements for providing at least a portion of the controlled heating of the first and second reservoirs (e.g. constituted by heater resistances placed against or integrated in the walls of the reservoirs), or

else a combination of the first and second electric heater elements.

Naturally, it is possible to envisage a third embodiment of the device of the invention in which the temperature regulation means comprise both a fluid circuit (as in the first embodiment) and electric heater elements (as in the second embodiment).

The device of the invention may include additional characteristics taken separately or in combination, and in particular:

- each of the first and second reservoirs may comprise a top portion and a bottom portion which are interconnected by a narrow intermediate portion, the link means communicating with the bottom bags, the top portions and the bottom portions each further including a leaktight inlet. Simultaneously, pressurization means may include a fluid pump for feeding pressurization fluid at high pressure via a (main) first portion of the pressurization circuit, and a second portion of the pressurization circuit connected to top and bottom pressure-regulating valves controlled by the control module for feeding each top and bottom reservoir portion via the leaktight inlets with pressurization fluid at high pressure or at low pressure or indeed at intermediate pressure. Under such circumstances, it is particularly advantageous for the first portion of the pressurization circuit to include a sub-portion immersed in the heat-conveying fluid (liquid or gas) which is received in the main container so as to feed the second portion of the pressurization circuit with pressurization fluid that has been heated. This makes it possible to minimize temperature disturbances to the culture fluid. Furthermore, it is also possible to provide an auxiliary container housed outside the main container in contact with the heat-conveying fluid (a "bain-marie"), containing a humidifying fluid and fed with fluid under pressure by the sub-portion of the first portion of the

pressurization circuit so as to feed the second portion of the pressurization circuit with a pressurization fluid presenting a selected degree of humidity. This is particularly important when the flexible bags are semi-permeable;

- at least two, and preferably three or four, wells may be placed in series and communicate with one another via link means, a first well being connected to the first reservoir while a well opposite to the first is connected to the second reservoir;

- the temperature regulation means may include at least one temperature sensor for providing the control module with measurements representative of the temperature inside a well, or in the immediate vicinity thereof; and

- a cover may be provided to isolate the wells from the outside, and possibly also to isolate the reservoirs and indeed the entire device.

The invention also provides an installation for culturing cells and tissue and comprising at least two devices of the above-described type placed in parallel and sharing a single control unit controlling all of their control units, or itself performing their functions.

This installation may include a main fluid circuit feeding the wells and/or the reservoirs of each device in parallel. In which case, it is particularly advantageous to provide central temperature regulation means controlled by the main control unit and serving to maintain a common selected first temperature or a common selected first temperature cycle within the wells of each device, and/or to place the culture fluid which flows out from at least one of the first and second reservoirs of each device to feed its wells at a common selected second temperature or at a common selected second temperature cycle.

In a variant, the main control unit controls the temperature regulation means of each device in such a manner as to cause them to maintain a first selected temperature or a first selected temperature cycle within the wells of the corresponding device independently of one another, and/or place the culture fluid which leaves at least one of the first and second reservoirs of the corresponding device to feed its wells at a second selected temperature or a second selected temperature cycle, independently of one another.

The installation may also include a main cover for isolating the wells of each device simultaneously from the outside, possibly together with the associated reservoirs or even the complete devices.

Other characteristics and advantages of the invention appear on examining the following detailed description and from the accompanying drawings, in which:

- Figure 1 is a fragmentary diagrammatic cross-section view of a culture device of the invention, having a plurality of chambers;

- Figures 2A and 2B are perspective views of two inner half-shells of the reservoirs of Figure 1, respectively before and after being assembled together;

- Figure 3 is a perspective view of the two inner half-shells of Figure 2 prior to being assembled together with two outer half-shells of the reservoirs;

- Figures 4A and 4B are perspective views showing how an inner half-shell is positioned inside the corresponding outer half-shell, respectively before and after assembly;

- Figure 5 is a diagrammatic perspective view of a culture installation constituted by four culture devices placed in parallel;

- Figure 6 is a perspective view of an assembly of four double-shell reservoirs for an installation of the type shown in Figure 5;

- Figure 7 is a diagram showing a sequence of eight successive go-and-return stages for culture fluid in a laminar type mode of flow;

- Figure 8 is a diagram showing a sequence of four successive go-and-return stages for culture fluid in a turbulent type mode of flow; and

- Figure 9 shows a variant of the turbulent mode shown in Figure 8.

The accompanying drawings are in essence definitive in nature. Consequently, they can serve not only to contribute to describing the invention, but they can also contribute to defining it, where appropriate.

Reference is made initially to Figures 1 to 4 for describing a cell and tissue culture device in a non-limiting embodiment.

The device 1 shown in Figure 1 comprises firstly a first reservoir 2 having a top portion 3 coupled to a bottom portion 4 via an intermediate portion 5. The reservoir 2 is defined by rigid walls 15 which give it a volume that is constant and which is discussed further below.

In the example shown, the top portion 3 of the reservoir houses a top flexible bag 6. Similarly, the bottom portion 4 houses a bottom flexible bag 7 which is connected to the top bag 6 via a duct 8 housed in the intermediate portion 5, being received closely therein so that the top and bottom portions 3 and 4 of the first reservoir 2 are isolated from each other.

The top bag 6 has an inlet/outlet 9 adapted so as to be capable of co-operating in leaktight manner with a top opening 10 formed in one of the partitions of the top portion 3 of the first reservoir 2. Thus, the top bag 6 may be connected to top access control means 11, themselves connected to a culture fluid or gas feed module, or as shown to a nutrient (or culture fluid) container 14, which is preferably pressurized. For reasons of compactness, the nutrient container 14 is

placed beneath the top portion 3 of the reservoir 2, however it could be located elsewhere.

Similarly, the bottom bag 7 has an inlet/outlet 12 adapted to co-operate with a leaktight opening formed in the wall of the bottom portion 4 of the first reservoir 2, or else as shown in Figure 1, so as to co-operate with access control means 13, in this case housed outside the bottom portion 4 of the reservoir 2.

The bottom bag 7 may comprise two substantially rigid membranes so as to prevent it from being completely flattened when it is subjected to very high pressures, since that would impede good flow of the culture fluid.

Also preferably, in the intermediate portion 5, the first reservoir 2 has an additional opening enabling a liquid or a gas to be injected into or extracted from the inside of the duct 8 either manually or automatically. The opening is preferably fitted with a septum, which is particularly suitable when the injection or extraction device is a syringe fitted with a needle. It is also preferable to provide a septum in each of the bottom and top portions of the reservoirs.

Also preferably, the top and bottom bags 6 and 7 are made of a porous material, at least in a material that is porous going from the outside towards the inside. They may be bags made of silicone, or of polydimethylsiloxane (PDSM), or indeed of polytetrafluoroethylene (PTFE), or indeed of dimethyl and methylvinyl siloxane polymers. This enables gas to be exchanged between the culture fluid which is inside the flexible bags and the gas which is trapped inside the top and bottom portions 3 and 4 of the first reservoir 2. These bags may be made of materials that are different so as to provide different functions, in particular concerning exchange with the fluid which is contained inside the reservoirs (generally the pressurization gas(es) described in greater detail below). In addition, the bags in a single reservoir may present shapes and volumes that are different.

In the example shown in Figure 1, the bottom bag 7 communicates with a culture well 18-1 to 18-3 via the access control means 13 and the bottom opening formed in the wall of the reservoir.

The access control means 13 are preferably of the "pinch" type. They have a hollow end into which one end of link means 20 is inserted, the link means being made in the form of a duct (or tube) having its opposite end opening out into the culture chamber 19-1 of the first well 18-1. This first culture chamber 19-1 communicates with the second culture chamber 19-2 housed in the second well 18-2 via another link means 21 likewise implemented in the form a duct (or tube). Similarly, the second culture chamber 19-2 communicates with the third culture chamber 19-3 housed in the third well 18-3 via another link means 21 made in the form of a duct (or tube). Finally, in this example, a last link means 20 provides communication between the third culture chamber 19-3 and a second reservoir 25, which is described below.

The second reservoir 25 is preferably substantially identical to the first reservoir 2 as described above with reference to Figures 1 to 4. Consequently, in this example, it comprises a top portion 26 having a top flexible bag 27 housed therein, a bottom portion 28 having a bottom flexible bag 29 housed therein, and a narrow intermediate portion 23 housing an intermediate duct 24 coupling the top bag 27 to the bottom bag 29. This duct 24 is likewise housed narrowly in the intermediate portion 23 so that the top portion 26 is isolated in terms of gas-tightness from the bottom portion 28.

The top bag 27 has a suitable inlet/outlet 30 connected to access control means 31 which, like the access control means 11, can be connected to a gas or fluid feed device 32 or to an extractor. Similarly, the bottom bag 29 has an inlet/outlet 33 which, in the example shown, is connected to access control means 34

located in this case outside the bottom portion 28 of the second reservoir 25.

The second reservoir 25 preferably also includes openings in its top, intermediate, and bottom portions 26, 23, and 28 enabling a liquid or a gas to be injected into or extracted from the pockets or the intermediate duct 24, either manually or automatically. These openings are preferably fitted with respective septums.

In this example, the access control means 34 are likewise preferably of the "pinch" type, having for this purpose a hollow end which is connected to the end of the link duct 20.

A circuit is thus established between the top bag 6 of the first reservoir 2 and the top bag 27 of the second reservoir 25 via the culture chambers 19-i (i = 1 to 3 in this example) and via the link means 20 and 21.

In order to enable culture growth to be controlled thermally, the device has temperature regulation means for regulating temperature inside the culture well(s), or for regulating the temperature of the culture fluid fed to the wells, or indeed, and preferably, for regulating temperature both in the wells and of the culture fluid, as shown in Figures 1 to 4.

In the embodiment shown in these figures, the temperature of the culture fluid is regulated in the two reservoirs 2 and 25 by circulating a heat-conveying fluid (liquid or gas) inside their rigid walls 15. More precisely, the walls 15 which define the top and bottom portions of the reservoirs 2 and 25 have fluid circulation spaces 35 integrated therein and forming part of a fluid circuit for heating purposes. As shown in Figures 2 to 4, it is advantageous for this circulation space 35 to be defined by assembling together an inner shell 16 and an outer shell 17 housing the inner shell 16.

The inner shell 16 is preferably constituted by assembling together two half-shells 16a and 16b which

define the top, intermediate, and bottom inner portions of each reservoir 2 and 25.

The outer shell 17 is likewise preferably constituted by assembling together two half-shells 17a and 17b having first holding means 36 (in this case orifices) for co-operating with second holding means 37 (in this case studs) formed on the outside surface of the inner shell 16. In its top portion it also has an inlet 42 fitted with a first connector 43 (suitable for connection to the "external" main portion of the heating fluid circuit), and in its bottom portion it has an outlet 44 provided with a second connector 45 suitable for being connected to a third or a fourth connector 46 fitted to the end wells 18-1 and 18-3. As a result, the heat-conveying fluid can circulate inside the walls 15 of the reservoirs 2 and 25 and provide effective temperature regulation for the culture fluid which circulates in the bags.

In order to provide temperature regulation in the wells, channels 47 are provided forming another portion of the heating fluid circuit. When the wells 18 are made in a thick solid block 48, the channels 47 are preferably formed by making hollows in said block 48 at the periphery of the zones defining the wells, and preferably also beneath them. In a variant, when the wells and the reservoirs are installed on a support plate, the support plate may include channels 47 for circulating a fraction of the heat-conveying fluid beneath the wells 18. The channels 47 are connected to one side of a third connector 46 for connecting to the second connector 45 of the first reservoir 2 and to the opposite side of the fourth connector 46 for connection to the first connector 43 of the second reservoir 25.

The heat-conveying fluid circulates in the main portion of the heating fluid circuit and thus reaches the walls 15 of the first reservoir 2 via the first connector 43, circulates in the inter-shell space 35, and then

reaches the second connector 45. It then penetrates into the channels 47 of the wells 18 via the third connector 46 and reaches the fourth connector 46. Thereafter it penetrates into the walls of the second reservoir 25 via its second connector 45, circulates in the inter-shell space, and then reaches its first connector 43 from which it returns to the main portion of the heating fluid circuit.

In order to enable the heat-conveying fluid to circulate, the main portion of the fluid circuit includes firstly a main container 49 containing a fraction of the heat-conveying fluid and including electric heater means 51, e.g. adjustable heater resistances, an inlet 52 connected via a duct 53 to the first connector 43 of the second reservoir 25, and an outlet 54 connected to a pump (not shown) which feeds the first connector 43 of the first reservoir 2 via another duct 55. This other duct 55 is preferably fitted with two parallel-connected solenoid valves (or pneumatic valves) for regulation purposes, 56 and 57, and with a pressure sensor 58 (or pressure contact). The temperature of the heat-conveying fluid in the main container 49 is selected in such a manner as to ensure that the culture fluid in the outlet 12 of the bottom bag 7 housed in the first reservoir 2 is at a temperature which is suitable for culture purposes.

Naturally, the temperature inside the wells can be different from or substantially identical to the temperature of the culture fluid leaving the first reservoir, depending on requirements.

In a variant embodiment, both reservoirs 2 and 25 and the wells 18 may be fed in parallel with the same heat-conveying fluid, or with heat-conveying fluids coming from two or three independent heating fluid circuits. It is also possible to provide a heating-fluid circuit for each portion of a reservoir. This makes it possible to provide reservoirs containing culture mediums placed at different temperatures on either side of the

culture chamber, so as to create temperature profiles or temperatures cycles.

The supplies (or nutrient container) 14 and/or the gas or fluid feed devices (or waste vessels) 32 may also possess their own thermostat circuits so as to maintain their respective contents at selected temperatures which might optionally be different. The thermostatically controlled temperatures may involve heating or cooling. It is generally preferable to maintain them at temperatures lying in the range about 3°C to about 12°C in order to ensure that the culture medium is stable.

In another variant, which is completely different, the temperature regulation means comprise electric heater means such as heater resistances or controlled temperature profile (CTP) elements. Such means may be placed at selected locations on or in the walls defining the reservoirs and/or the wells.

It is also possible to envisage combining heater resistances and a heating fluid circuit.

The power of the electric heater means and/or the flow rate of the heat-conveying fluid is/are controlled by a control unit 50 so as to govern the temperature of the heat-conveying fluid.

Furthermore, in order to improve temperature control in the wells and/or in the reservoirs, one or more temperature sensors may be provided at selected locations to deliver temperature measurements to the control unit.

In order to govern the inside volumes of the top bags 6 and 27 and of the bottom bags 7 and 29, the device of the invention includes pressurization means which are described below with reference to Figure 1.

In the embodiment shown, pressurization means are used which are common to two reservoirs 2 and 25, which means are housed in an external unit 105 (such as the unit represented by dashed lines in Figure 1) together with most of the temperature regulation means. However, in a variant, each reservoir could have its own

pressurization means housed in respective units placed, for example, beneath the top portions of the reservoirs.

The pressurization means comprise a high pressure pressurization circuit 59 having a pressure booster (or pump) 60 fed with ambient air 61 and feeding a pressurized supply 62, preferably coupled to a pressure sensor (or pressure contact) 63. The reserve 62 feeds a main duct 64 fitted with a pressure regulator 65 and then a first flow rate regulator 66 and a particle filter 67 (e.g. having a 0.01 micron grid). When the device is for use with a plurality of different pressurization fluids (e.g. air and carbon dioxide), an auxiliary duct 68 is provided which is fed with auxiliary fluid(s) 69 (e.g. carbon dioxide), having a second pressure regulator 70 followed by a second flow rate regulator 71 and feeding the main duct 64 between the first pressure regulator 65 and the filter 67. Under such circumstances, it is advantageous to provide a third flow rate regulator 72 between the filter 67 and the point where the auxiliary duct 68 is connected.

The main duct 64 serves to feed pressurized fluid to the two reservoirs 2 and 25 and also to the culture fluid container 14. In order to minimize temperature disturbances which might be generated by the pressurization fluid on penetrating into the top and bottom portions of the reservoirs 2 and 25, it is heated by means of the heat-conveying fluid that is located in the main container 49. To do this, a portion 73 of the main duct 64 is housed in the main container 49, preferably in the form of a coil therein or in any form that encourages heat exchange.

In addition, in order to be able to control the humidity of the pressurization fluid before it penetrates into the reservoirs 2 and 25, an auxiliary container 74 is preferably provided in the main container 49 and partially filled with a humidifying liquid, the portion 73 of the main duct that is immersed in the heat-

conveying fluid opening out into said auxiliary container.

The portion 75 of the main duct 65 which opens out into the auxiliary container 74 feeds, preferably via a thermometer-hygrometer 76, firstly a first port 77 at high pressure (e.g. about 45 millibars (mbar)) which is fitted with four valves 78, 79, 80, and 81 connected in parallel, secondly a second port 82 at high pressure (e.g. about 45 mbar) which opens out into the culture fluid container 14, thirdly a third port 83 at low pressure (e.g. about 10 mbar) which is fitted with four valves 84, 85, 86, and 87 connected in parallel, preferably together with a fourth flow rate regulator 88 placed upstream from the valves, and fourthly a fourth port 89 at intermediate pressure (e.g. about 30 mbar) which is preferably fitted with a fifth flow rate regulator 90 followed by a solenoid valve 91 (or a pneumatic valve) and a pressurized supply 92 feeding in parallel the four valves 78, 79, 80, and 81 which are preferably solenoid valves or pneumatic valves.

In a variant, pressurization fluid circuits may be provided that are different in order to govern the volumes of the bags housed inside the top and bottom portions of the same reservoir. This can make it possible to use different pressurization fluids within the same reservoir so that the bags perform different functions, for example in order to perform comparative tests.

The various solenoid valves (or pneumatic valves) 78-81 and 84-87 are preferably all of the three-port type (two inlets and one outlet), the outlets of the solenoid valves (or pneumatic valves) 78-81 feeding respective ones of the inlets of the solenoid valves (or pneumatic valves) 84-87 whose outlets act respectively via connectors 93-96 connected to the connectors 39, 41 installed in the leaktight inlets 38, 40 to feed the insides of the top and bottom portions 3 and 4 of the first reservoir 2 and of the top and bottom portions 26

and 28 of the second reservoir 25 so as to govern the volumes of the flexible bags contained therein.

These solenoid valves (or pneumatic valves) may also be used for governing the states of the access control means 11, 13, 31, and 34 of the wells 18 and the flexible bags, which, as mentioned above, are preferably of the "pinch" type and are, for example, as described in patent document FR 00/00548. However that is merely one possibility amongst others, and switches or valves could also be used.

All of the solenoid valves and the pressurization fluid pumps are controlled by the electronic control unit 50 which is provided for this purpose with microprocessors (or a microcontroller) mounted on a card which is preferably connected to a link interface 97 (e.g. of the RS232 type) in order to enable it to be remotely controlled by a process computer.

Once programmed, the microcontroller 50 controls the solenoid valves (or pneumatic valves) in such a manner as to apply high and/or low pressure sequences to the bags by means of the pressurization fluid, in accordance with the requirements and in the top and bottom portions 3 and 4 of the reservoirs 2 and 25. Naturally, the microcontroller 50 may include a memory 98, preferably a re-writable memory, containing a multiplicity of culture programs, each culture program defining first and second pressure sequences for governing the respective volumes of the various flexible bags, and also the regulated temperatures of the wells and/or of the heat-conveying fluid.

As mentioned above, instead of using a microcontroller for governing a single pressurization circuit, it is possible to use the same microcontroller to govern two pressurization circuits that are at least partially independent, e.g. installed beneath the top portions of the reservoirs. In another variant, it is possible to use two independent microcontrollers that

have previously been synchronized in order to govern two independent pressurization circuits.

The device preferably includes a cover for isolating the well(s) and possibly also the reservoirs from the external medium. This serves not only to avoid exchanges taking place through the various septums, but also to limit temperature disturbances. This also serves to establish a "mechanical" protective barrier around the wells. The cover can also cover the entire device, thereby forming an enclosure defining a biological barrier which is particularly useful when said device is not itself placed under a laminar flow hood. The shape of the cover and the material from which it is made can be selected so as to enable the cells and tissue contained in the wells to be observed under a microscope or using any other suitable optical means while they are being cultured. For this purpose, the cover is preferably made of a material that is not breakable, and that is transparent over the wells.

An outlet for connection to atmospheric pressure may also be provided in the top and bottom portions of the reservoirs 2, 25 being fitted with a solenoid valve (or a pneumatic valve) 99-102 under the control of the control unit 50. In addition, as shown in Figures 2 to 4, the inlets 9, 30 of the top flexible bags 6, 27 are preferably placed in a rigid duct 103 defined by the rigid walls of the inner half-shells 16a and 16b and are provided with respective top cavities 104 fitted with draining means (not shown) so as to evacuate any microbubbles of air that might form in operation in the flexible bags of the reservoirs 2 and 25.

The device of the invention can be considered as comprising a control unit coupled with "consumable" type elements (reservoirs and/or wells) that are possibly for single use only. This can be achieved merely by providing the outer control, pressurization, and temperature regulation unit 105 with first and second connection

means 93-96, 106 connected respectively to the pressurization and temperature regulation circuits, and secondly the two reservoirs 2 and 25 of each device with third and fourth connection means 39, 41, and 43 respectively connected to the top and bottom inside portions of the reservoirs and to the inter-shell space 35, and then connect the first connection means 93-96 to the third connection means 39, 41 and the second connection means 106 to the fourth connection means 43.

In order to start a new culture, the used consumables are disconnected (the reservoirs and/or wells) and they are replaced with new consumables which are connected to the external control unit.

As shown diagrammatically in Figures 5 and 6, it is possible to place a multiplicity of devices 1 in parallel so as to constitute an installation for culturing cells and tissue, either for high throughput (identical cultures) or else for a high degree of differentiation (with different cultures). In this example, the installation has four parallel devices 1-1 to 1-4, each device 1-i (in this case i = 1 to 4) having three culture wells 18-j (in this case j = 1 to 3) connected in series. The reservoirs with respective heat-conveying fluid circulation spaces are connected to one another, for example by fitting studs 37 on the inner half-shells 16b through suitable holes 108 formed in the outer half-shells 17a and 17b (see Figure 6).

These devices can be completely independent from one another. Under such circumstances, they may either have a common control unit which controls pressurization and temperature regulation circuits that are independent from one another, or else independent control units each controlling a single pressurization circuit and a single temperature regulation circuit. Under such circumstances, the regulation temperatures and/or the pressurization fluids can differ from one device to another. However

such devices may also depend on one another because some of their wells may be in communication.

It is also possible to envisage an installation in which the devices have wells that are independent from one another, sharing a common pressurization circuit and a common temperature regulation circuit controlled by a common (or main) control unit. Under such circumstances, the major portion of the pressurization means and of the temperature regulation means, and also the main control unit are housed in an external unit 105 (such as that represented by dashed lines in Figure 1). As a result, it is possible to form an installation in which the devices constitute modular elements of the "consumable" type, possibly for single use only. This can be achieved merely by providing the outer control, pressurization, and temperature regulation unit 105 with first and second connection means 93-96, 106 connected respectively to the pressurization and temperature regulation circuits, and secondly the two reservoirs 2 and 25 of each device with third and fourth connection means 39, 41, and 43 respectively connected to the top and bottom inside portions of the reservoirs and to the inter-shell space 35, and then connect the first connection means 93-96 to the third connection means 39, 41 and the second connection means 106 to the fourth connection means 43.

To proceed with new cultures, the used consumables (reservoirs and/or wells) are removed and replaced by new consumables whose wells have optionally been inoculated with cells.

In such an installation, the number of devices connected in parallel can vary depending on requirements.

In an installation of the invention, as in a device of the invention, the culture wells 18-j may be connected in series on a support plate 107 as shown in Figure 5, or else they may be formed directly by hollowing out a thick solid block 48 (as shown in Figure 1).

In the first example (Figure 5), the support plate 107 may have housings for receiving each of the wells 18-i-j (in this case i = 1 to 4 and j = 1 to 3) and channels 47 for circulating a fraction of the heat-conveying fluid close to the peripheries of the wells. The support plate 107 may also have channels or ducts for circulating a fraction of the pressurization fluid. In the second embodiment, the culture wells of the devices may be made in independent solid blocks or in a single block. Details concerning embodiments of wells suitable for use in a device of the invention are given in patent document FR 00/00548.

As mentioned above when describing the device 1, it is advantageous to provide a main cover so as to isolate the wells from the outside, and possibly also the two reservoirs of each device of the installation, or indeed all of the devices. This makes it possible to avoid using respective covers for each of the devices.

Examples of implementation (in other words first and second sequences of pressures for governing the volumes of the bags of the reservoirs) of the device and the installation of the invention are to be found in patent document FR 00/00548. It is merely recalled herein that the installation and the device are suitable for operating in the various modes mentioned below.

A "laminar" mode consists in causing the culture fluid to rise into the top bag of one of the two reservoirs so as to establish a difference in height between the top bag and the bottom bags of the two reservoirs, and then in allowing the culture fluid to flow under gravity from the top reservoir towards the bottom reservoirs, and cause the culture fluid to rise towards the top bag of the other reservoir. The same operations are then repeated in the opposite direction (the "return direction") in order to perform one complete cycle ("go-and-return") between the two reservoirs via the wells. The number of cycles is selected as a function

of the type of culture to be performed in the wells 18. The four steps of a go-and-return cycle in laminar mode are grouped together in Figure 7. The number of successive cycles is selected as a function of the type of culture to be performed.

A "turbulent" mode consists in applying high pressure continuously to the bottom bags 7 and 29 of the first and second reservoirs 2 and 25. In other words, the first and second sequences of the top bags of the first and second reservoirs are constituted by a succession of four low pressure periods. This mode has only two steps which are grouped together in the form of a "go-and-return" cycle in Figure 8. The number of successive cycles is selected as a function of the type of culture performed. This mode may be implemented in a first variant (Figure 9) in which the high pressure is not maintained continuously on the two bottom bags 7 and 29, but on the two top bags 6 and 27. This enables culture fluid to be caused to flow very quickly between the two bottom bags 7 and 29, given that said fluid can no longer rise because of the high pressures in the top bags 6 and 27. In a second variant (not shown), the first sequence applied to each bag of the first reservoir consists in alternating first periods of high pressure with second periods of low pressure, and the second sequence applied to each bag of the second reservoir consists in alternating first periods of low pressure and second periods of high pressure.

The two above-described modes of operation, laminar and turbulent, and also the variant mode, are merely a few of the numerous examples that can be envisaged. Thus, it is possible to combine turbulent operation cycles with laminar operation cycles.

The invention applies to very many types of cells and tissue, such as, in particular:

- cells of the intestine: intestine 407, Caco-2, Colo 205, T84, SW 1116, WiDr, HT 29, HT 115, HT 55;

- endothelial cells: human aortic smooth muscle cells (HAOSMC);
- epidermal cells: human epidermal keratinocyte neonatal (NHEK-Neopooled), Equine Dermis;
- cancer cells: HeLa, CHO-K1;
- intestine type fibroblast cells: CCD-18Co;
- fibroblast cells of MRC-5, 3T3, Wi-38 type;
- myelomas: SP20-Ag14, P3X63 Ag8 653, MPC11;
- hybridomas;
- insect cells: SF9.

This list is not exhaustive in any way; it merely gives examples.

The invention is not limited to the modes of operating the device and the installation as described above merely by way of example, and on the contrary covers all variants that the person skilled in the art might imagine within the ambit of the following claims.

Thus, in the above a temperature regulation circuit is described in which a heat-conveying fluid is circulated for the purpose of raising temperature. However, it is possible to make use also of an auxiliary temperature regulation circuit in which the heat-conveying fluid that circulates serves to remove heat in order to refrigerate certain media, for example the reserves. Naturally, under such circumstances, the device of the invention needs to be fitted with cooling means under the control of the control module.

Furthermore, in the description above, the wells are placed at first selected temperatures and/or the fluid(s) and one or more second selected temperatures. However, it is possible to envisage placing the well(s) under first temperature cycle(s) or profile(s) and/or the fluid(s) under second temperature cycle(s) or profile(s).

In addition, it is also possible to regulate the inlet section of each reservoir and of the chamber, particularly when they are fed by a common heat-conveying

circuit, so as to control their respective temperatures independently.

Finally, the temperature regulation means may be arranged in such a manner as to impart a thermal shock to the inside of the chamber and/or the wells. This can be particularly advantageous when it is necessary to modify the state of cell membranes. The thermal shock may be combined with a change in pressure achieved by controlling the flow rate of the fluid and/or the internal pressure of the chamber.

CLAIMS

1. A device for culturing cells and tissue, the device being of the type comprising at least one culture well (18-i) arranged to define a chamber (19-i) suitable for receiving cells or tissue to be cultured, first and second reservoirs (2 and 25) each housing at least one flexible bag (6, 7; 27, 29), at least one of the bags of the reservoirs being suitable for receiving a culture fluid, link means (20, 21) coupled to said well and to said bags to enable the culture fluid to flow from one reservoir to the other via said well, pressurization means (60-92) arranged to apply to the bags of the first and second reservoirs (2 and 25) respective first and/or second sequences of external pressures defined by at least one control module (50) for causing the culture fluid to flow through said well, the device being characterized in that it includes temperature regulation means (35, 47, 49, 51-58) controlled by said control module and arranged to maintain a first selected temperature or a first selected temperature cycle inside said well (18-i) and/or to subject the culture fluid leaving at least one of said first and second reservoirs (2 and 25) in order to feed said well to a second selected temperature or to a second selected temperature cycle.

2. A device according to claim 1, characterized in that said temperature regulation means comprise a fluid circuit including a first portion (47) integrated in the walls defining said well (18-i) and arranged to enable a heat-conveying fluid to circulate therethrough.

3. A device according to claim 2, characterized in that the first portion of the fluid circuit includes first connection means (46) opening out into circulation

channels (47) for the heat-conveying fluid that are integrated in the walls of the well (18-i).

4. A device according to any one of claims 1 to 3, characterized in that said temperature regulation means comprise a fluid circuit including second and third portions (35) respectively integrated in the walls (15) defining the first and second reservoirs (2 and 25) and arranged to allow a heat-conveying fluid to circulate therethrough.

5. A device according to claim 4, characterized in that each of the first and second reservoirs (2 and 25) is made by assembling together an inner shell (16) and an outer shell (17) housing the inner shell and defining an inter-shell space (35) in which the heat-conveying fluid can circulate.

6. A device according to claim 2 or claim 3 combined with claim 4 or claim 5, characterized in that the second portion (35) of the fluid circuit is arranged to feed heat-conveying fluid to the first portion (47), and the third portion of said fluid circuit (35) is arranged to collect the heat-conveying fluid that has circulated through the first portion (47).

7. A device according to claims 3, 5, and 6 in combination, characterized in that the second and third portions (35) of the fluid circuit include second and third connection means (43, 45) opening out into the inter-shell space (35) and suitable for being connected for a first sub-portion to the first connection means (46) and for a second sub-portion to a fourth portion (53, 55) of the fluid circuit for feeding and collecting heat-conveying fluid.

8. A device according to any one of claims 2 to 7, characterized in that said fluid circuit includes a pump coupled to a main container (49) containing a fraction of the heat-conveying fluid and electric heater means (51) for heating said heat-conveying fluid in controlled manner before it is fed to the first, second, and third portions (47, 43, and 45).

9. A device according to any one of claims 1 to 8, characterized in that said temperature regulation means comprise first and second electric heater elements for providing at least some of the controlled heating of the well.

10. A device according to any one of claims 1 to 9, characterized in that said temperature regulation means include second electric heater elements for providing at least a portion of the controlled heating of the first and second reservoirs.

11. A device according to claim 9 or claim 10, characterized in that said electric heater elements comprise heater resistances secured to walls defining the reservoirs and/or the well.

12. A device according to any one of claims 1 to 11, characterized in that each of the first and second reservoirs (2 and 25) comprises a top portion (3, 26) and a bottom portion (4, 28) interconnected via a narrow intermediate portion (5, 23), each top and bottom portion of the first and second reservoirs housing a respective flexible bag, said top and bottom flexible bags (6, 27; 7, 29) communicating with each other via the intermediate portions (5, 23), and said link means (20, 21) communicating with the bottom bags (6, 29), and the top and bottom portions (3, 26; 4, 28) of the first and second reservoirs (2 and 25) each further including a

leaktight inlet (38, 40), and in that the pressurization means comprise a fluid pump (60) suitable for feeding high pressure pressurization fluid via a first portion of the pressurization circuit (64) to a second portion of the pressurization circuit (77, 82, 83, 89) that is connected to top and bottom valves (78, 81, 84, 87; 79, 80, 85, 86) controlled by the control module (50) and suitable for feeding each top and bottom portions (3, 26; 4, 28) of the first and second reservoirs via said leaktight inlets (38, 40) with pressurization fluid at a pressure that is high, low, or intermediate.

13. A device according to claims 8 and 12 in combination, characterized in that the first portion of the pressurization circuit (64, 65) includes a sub-portion (73) immersed in the heat-conveying fluid contained in the main container (49) in such a manner as to feed the second portion of the pressurization circuit (77, 82, 83, 89) with heated pressurization fluid.

14. A device according to claim 13, characterized in that said pressurization means include an auxiliary container (74) placed in said main container (49) in contact with the heat-conveying fluid, the container containing a humidifying fluid and being fed with pressurization fluid by the sub-portion (74) of the first portion of the pressurization circuit so that the pressurization fluid which feeds the second portion of the pressurization circuit (65) presents a selected degree of humidity.

15. A device according to any one of claims 1 to 14, characterized in that it includes at least two wells (18-i) placed in series and communicating with each other via said link means (21), one of the wells (18-1) being connected to said first reservoir (2) and the other well (18-3) being connected to said second reservoir (25).

16. A device according to claim 15, characterized in that it includes a third well (18-2) placed in series between the other two wells and communicating with each of them via said link means (23).

17. A device according to any one of claims 1 to 16, characterized in that the control module (50) includes a memory (98) of the re-writable type suitable for storing said pressure sequences and said first and second selected temperatures, or the first and second selected temperature cycles.

18. A device according to any one of claims 1 to 17, characterized in that said control module (50) is arranged to control the inlet sections of each reservoir and of the chamber when they are fed by a common heat-conveying circuit, so as to control their respective temperatures independently.

19. A device according to any one of claims 1 to 18, characterized in that it includes at least one nutrient container (14) and gas or fluid feed devices (32) connected to thermostat circuits, and in that said control module (50) is arranged to control said thermostat circuits in such a manner as to maintain the respective contents of the nutrient container (14) and/or of the gas feed devices (32) at selected temperatures.

20. A device according to any one of claims 1 to 19, characterized in that each reservoir portion is connected to a heating fluid circuit so that the portions of the reservoirs that are placed on either side of the chamber can be placed at different temperatures so as to create temperature cycles or profiles in said wells.

21. A device according to any one of claims 1 to 20, characterized in that said temperature regulation means

are arranged to impart a temperature shock to the inside of the chamber and/or the wells.

22. A device according to any one of claims 1 to 21, characterized in that the temperature regulation means include at least one temperature sensor suitable for supplying the control module with measurements representative of the temperature inside a well.

23. A device according to any one of claims 1 to 22, characterized in that it includes a cover for isolating at least the wells from the outside.

24. A device according to claim 23, characterized in that the cover is for isolating both the wells and the reservoirs from the outside.

25. An installation for culturing cells and tissue, characterized in that it comprises at least two devices (1-i) according to any one of claims 1 to 24 placed in parallel, together with a main control unit (50) for controlling said devices together.

26. An installation according to claim 25, characterized in that it includes a main fluid circuit feeding the wells and/or reservoirs of each device in parallel.

27. An installation according to claim 25, characterized in that it includes central temperature regulation means controlled by said main control unit (50) and arranged to maintain the same selected first temperature or the same selected first temperature cycle inside the wells of each device and/or for subjecting the culture fluid that leaves at least one of said first and second reservoirs of each device for feeding its wells to the same selected second temperature or to a same selected second temperature cycle.

28. An installation according to claim 27, characterized in that said main control unit (50) controls the temperature regulation means of each device so that they maintain independently of one another a first selected temperature or a first selected temperature cycle within the wells of the device and/or so that they act independently of one another to subject the culture fluid that leaves at least one of said first and second reservoirs of the device for feeding its wells to a second selected temperature or to a second selected temperature cycle.

29. An installation according to any one of claims 25 to 28, characterized in that it includes a main cover for isolating at least the wells of each device simultaneously from the outside.

30. An installation according to claim 29, characterized in that said main cover serves to isolate the wells and the reservoirs of each device simultaneously from the outside.

A B S T R A C T

A CELL AND TISSUE CULTURE DEVICE WITH TEMPERATURE REGULATION

A device for culturing cells and tissue comprises at least one culture well (18-i), first and second reservoirs (2 and 25) each housing at least one flexible bag (6, 7; 27, 29), at least one of which is suitable for receiving a culture fluid, link means (20, 21) coupled to the wells and to the bags to allow culture fluid to flow from one reservoir to the other via the well, pressurization means (60-92) subjecting the first and second reservoirs (2 and 25) respectively to first and/or second sequences of external pressures defined by a control module (50) for the purpose of governing the flow of culture fluid in the well, and temperature regulation means (49, 51-58) controlled by the control module and arranged to maintain a first selected temperature inside the well and to subject the culture fluid leaving the first and second reservoirs in order to feed the well to a second selected temperature.

Translation of the title and the abstract as they were when originally filed by the Applicant. No account has been taken of any changes that may have been made subsequently by the PCT Authorities acting ex officio, e.g. under PCT Rules 37.2, 38.2, and/or 48.3.

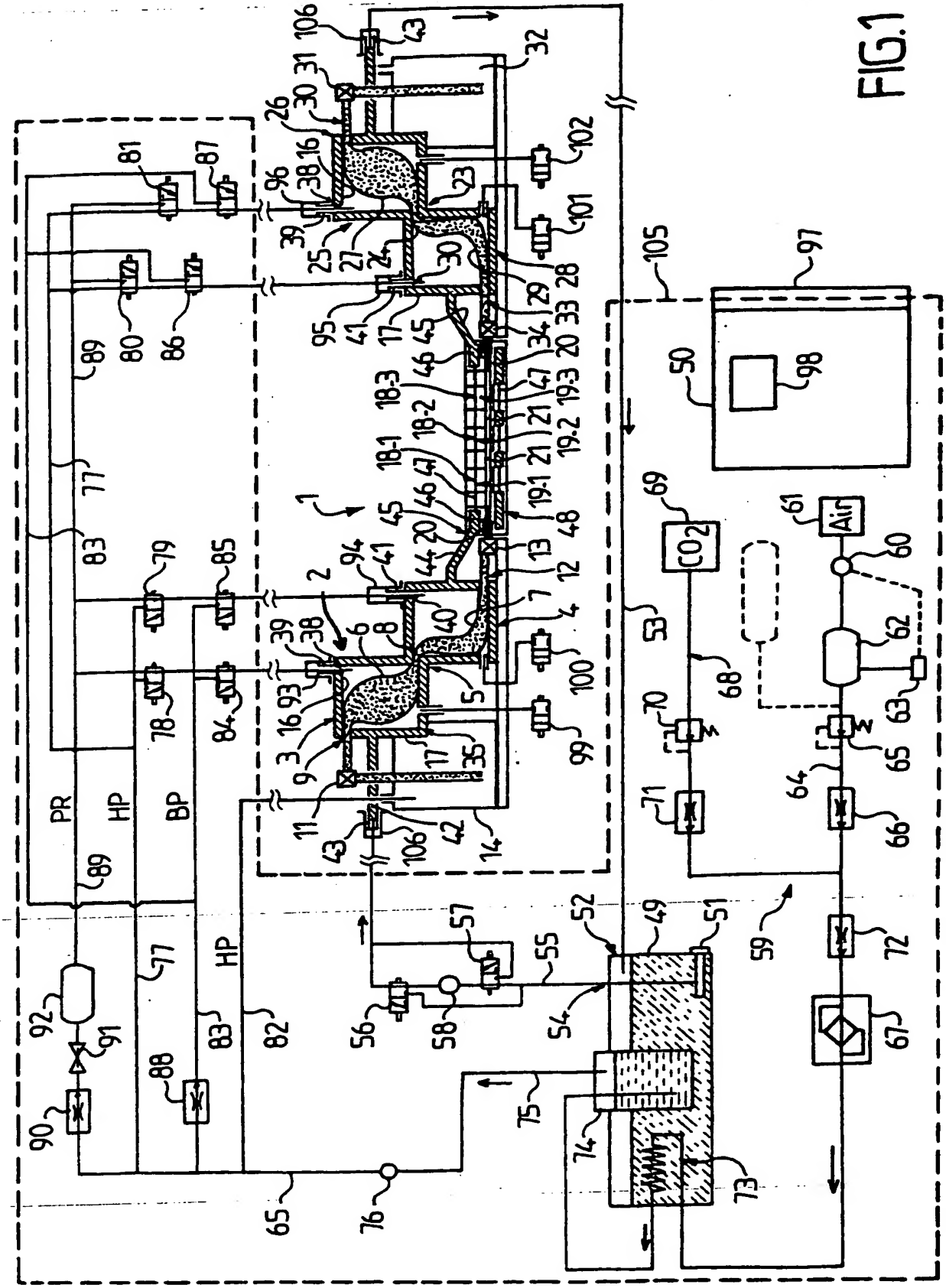

FIG.1

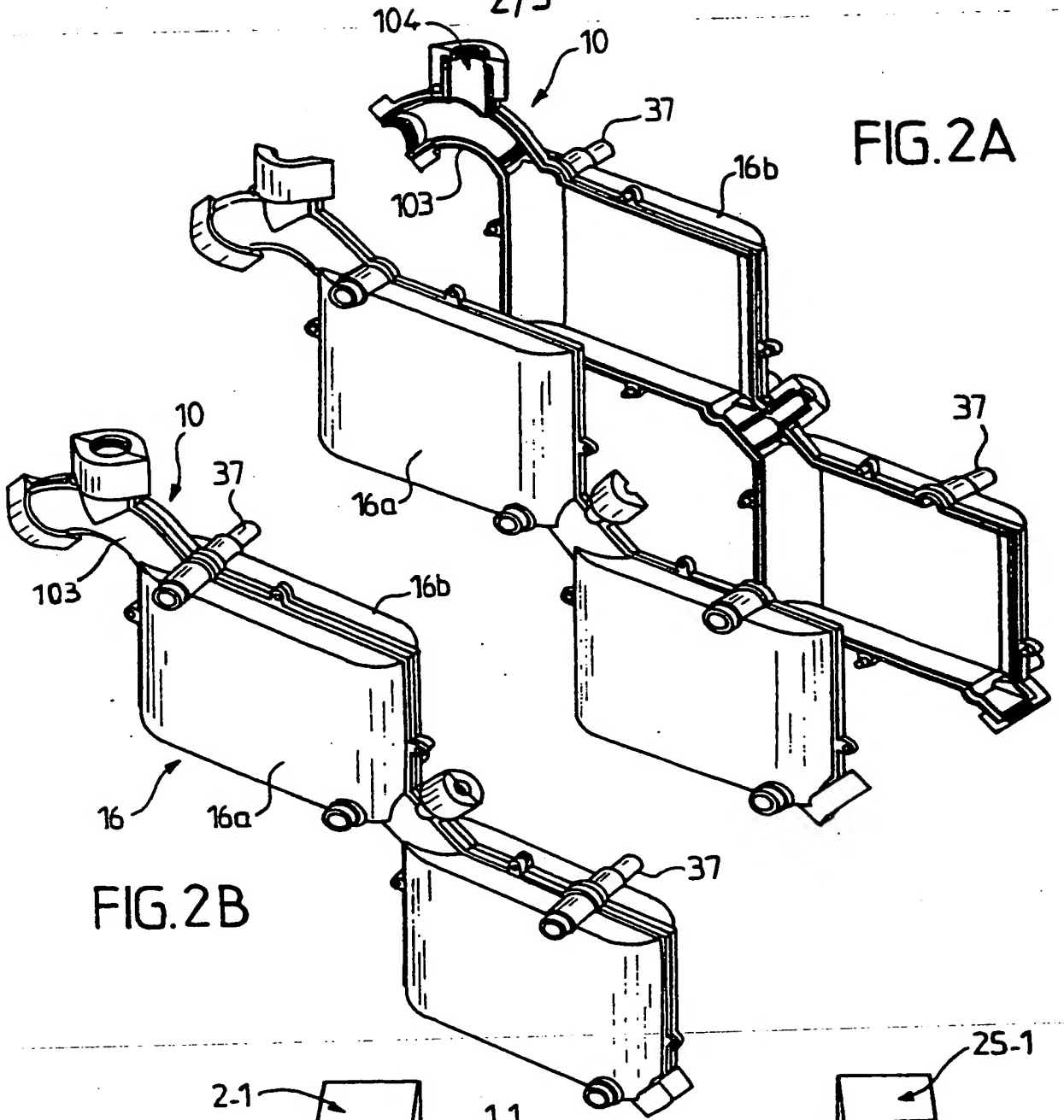

FIG.2A

FIG.2B

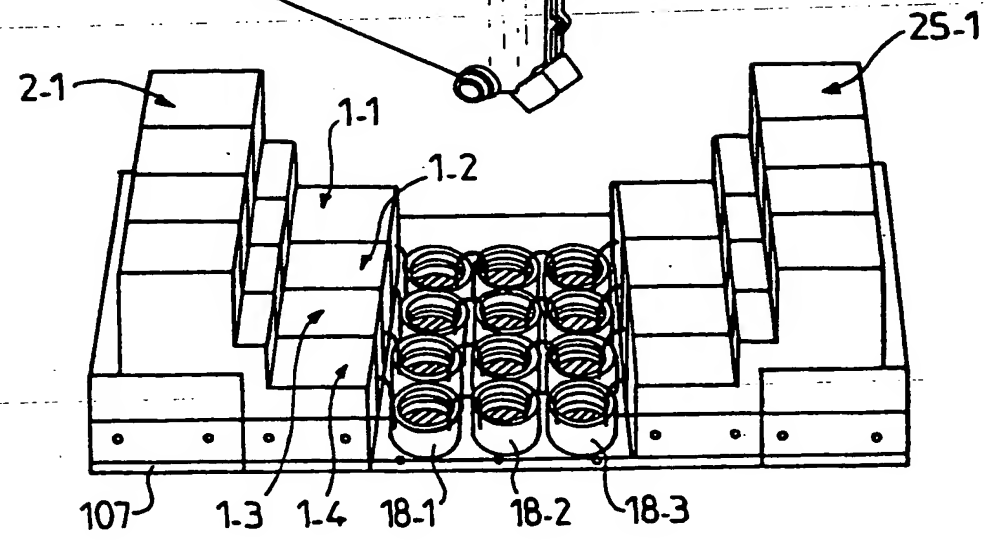

FIG.5

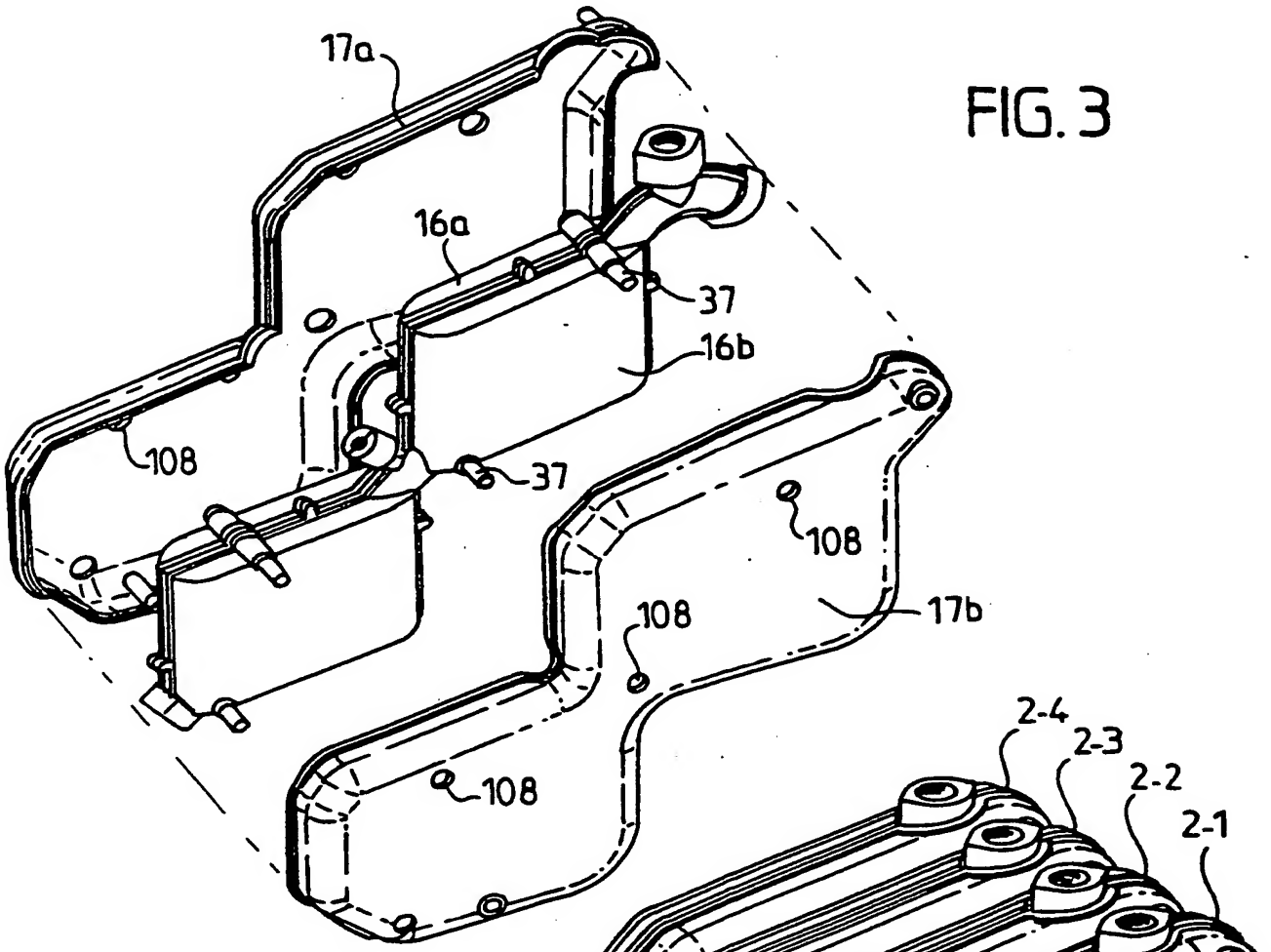

FIG. 3

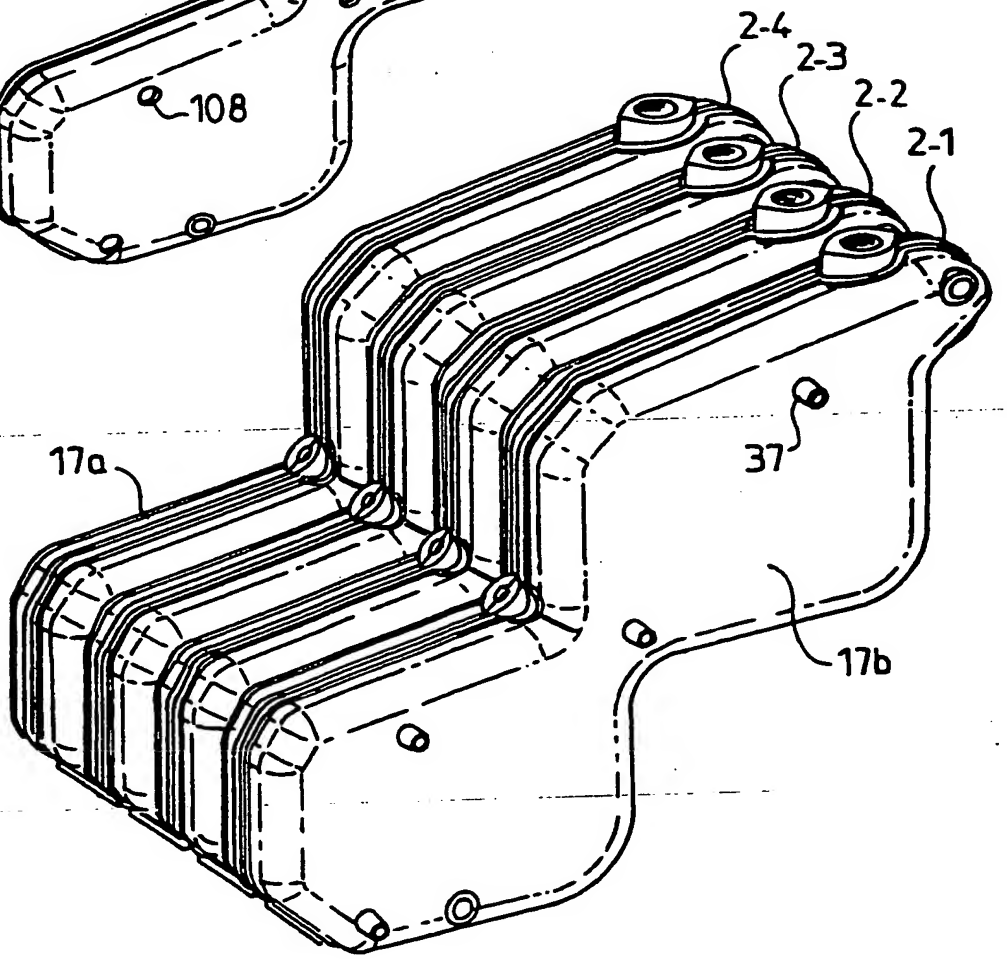

FIG.6

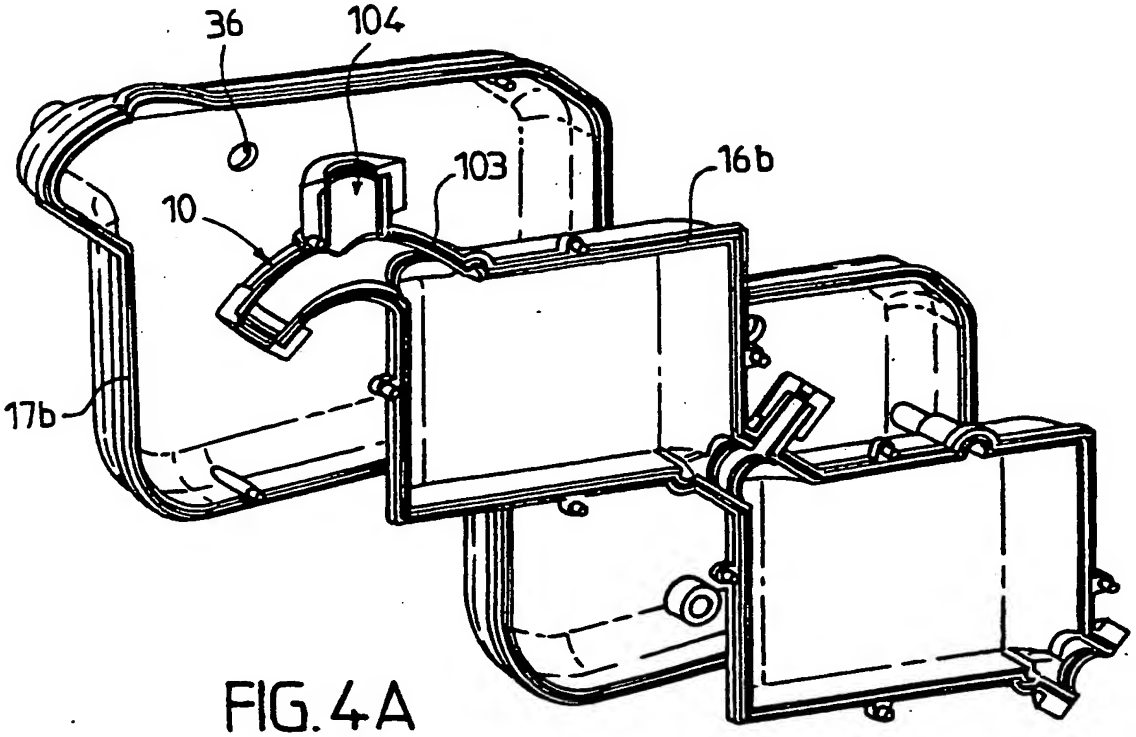

FIG.4A

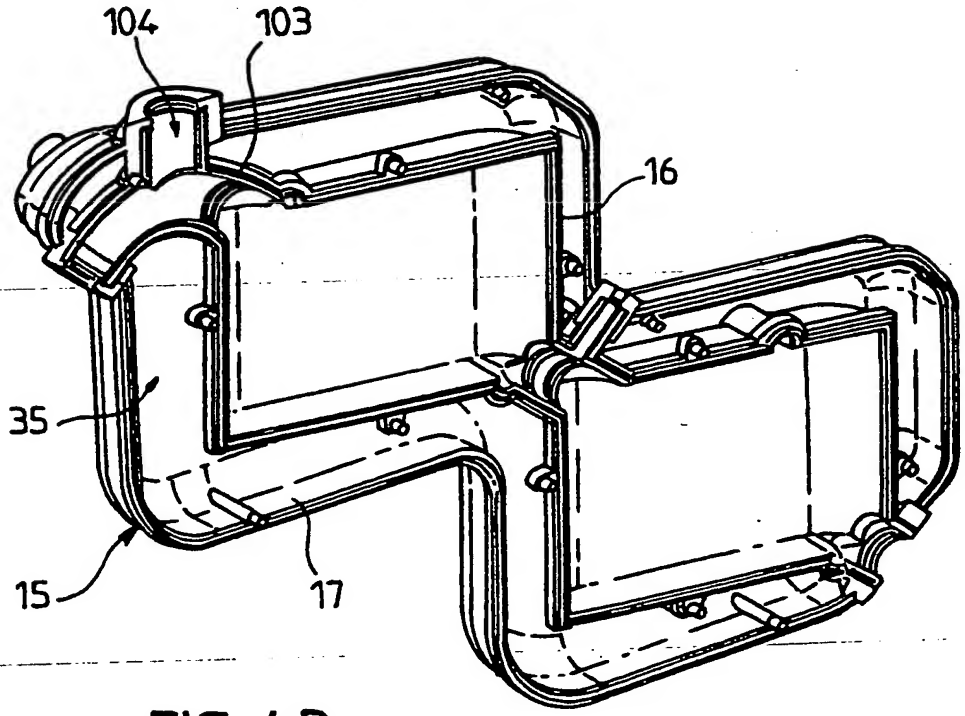

FIG.4B

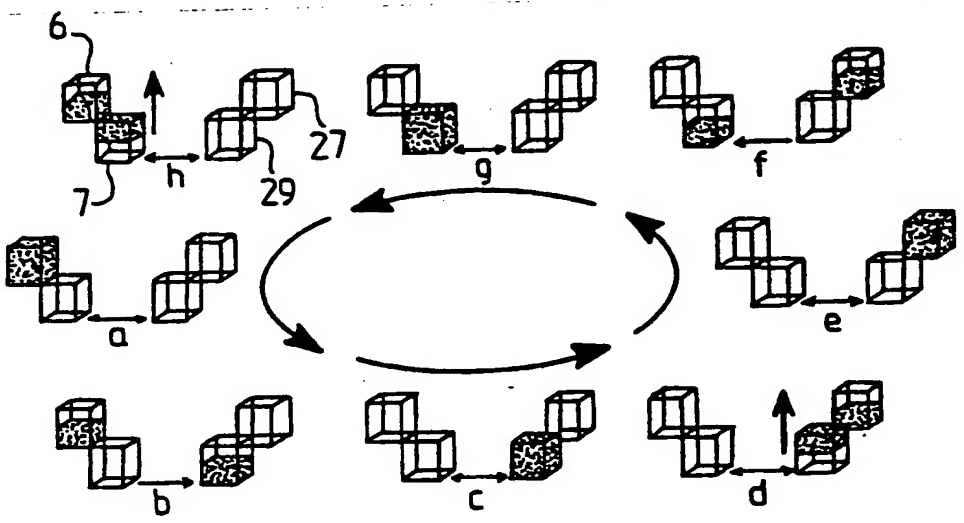

FIG.7

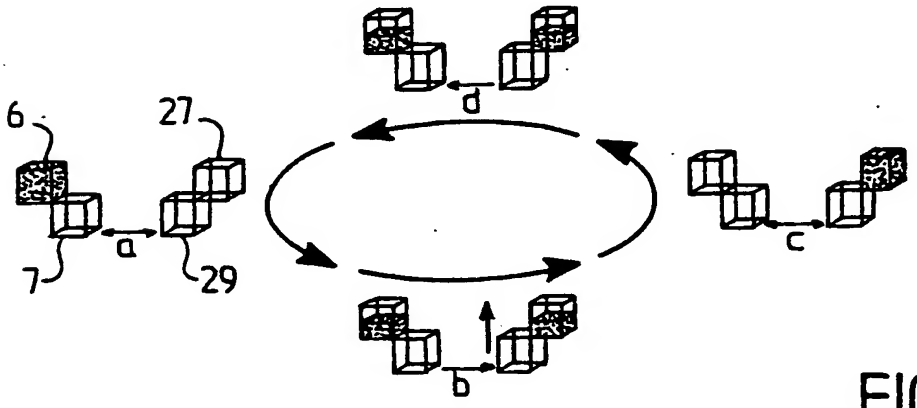

FIG.8

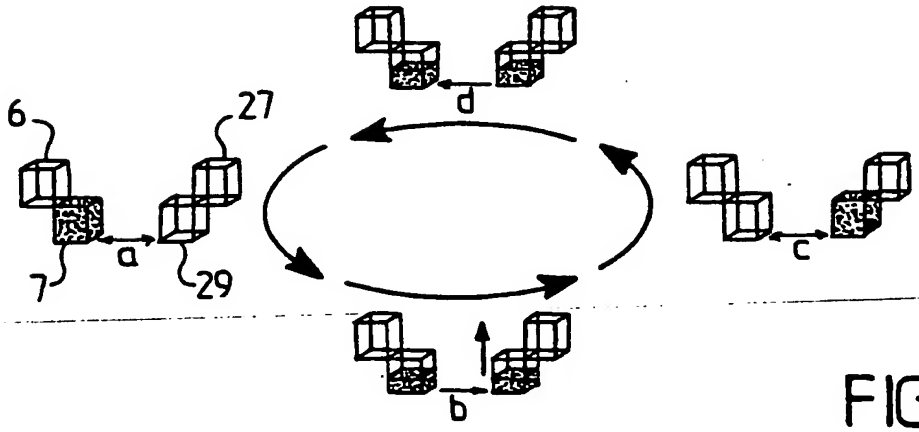

FIG.9